



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification ⁶ : A01N 43/04, A61K 31/70, C07H 21/02, 21/04, C12N 15/00, 15/09, 15/63, 15/70, 15/74, A61N 1/04	A1	(11) International Publication Number: WO 98/02040 (43) International Publication Date: 22 January 1998 (22.01.98)
(21) International Application Number: PCT/US97/05556 (22) International Filing Date: 4 April 1997 (04.04.97) (30) Priority Data: 08/682,433 17 July 1996 (17.07.96) US (71) Applicant: MEDTRONIC, INC. [US/US]; 7000 Central Avenue N.E., Minneapolis, MN 55432 (US). (72) Inventors: STOKES, Kenneth, B.; 17581 Eidelweiss Street N.W., Anoka, MN 55304 (US). MORISSETTE, Josée; Apartment 306, 1101 Paul Parkway, Blaine, MN 55434 (US). (74) Agents: PRESTON, Albert, W., Jr. et al.; Woodcock, Washburn, Kurtz, Mackiewicz & Norris LLP, 46th floor, One Liberty Place, Philadelphia, PA 19103 (US).		(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, UZ, VN, YU, ARIPO patent (GH, KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i>
(54) Title: SYSTEM FOR ENHANCING CARDIAC SIGNAL SENSING BY CARDIAC PACEMAKERS THROUGH GENETIC TREATMENT (57) Abstract The present invention provides delivery systems for delivering ion channel protein genetic material to cardiac cells in areas adjacent to where an electrode is to be positioned in a patient's heart to improve or correct the signal to noise ratio of cardiac signals, such as the P-wave. More specifically, there is provided a system for delivering sodium ion channel proteins or nucleic acid molecules encoding sodium ion channel proteins to a site in the heart adjacent to an electrode to increase the expression of the same, thereby enhancing the cardiac signal amplitude and enabling improved sensing of cardiac signals by an implanted pacemaker.		

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Swaziland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BA	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Barbados	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece	ML	Mali	TR	Turkey
BG	Bulgaria	HU	Hungary	MN	Mongolia	TT	Trinidad and Tobago
BJ	Benin	IE	Ireland	MR	Mauritania	UA	Ukraine
BR	Brazil	IL	Israel	MW	Malawi	UG	Uganda
BY	Belarus	IS	Iceland	MX	Mexico	US	United States of America
CA	Canada	IT	Italy	NE	Niger	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NL	Netherlands	VN	Viet Nam
CG	Congo	KE	Kenya	NO	Norway	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NZ	New Zealand	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	PL	Poland		
CM	Cameroon	KR	Republic of Korea	PT	Portugal		
CN	China	KZ	Kazakhstan	RO	Romania		
CU	Cuba	LC	Saint Lucia	RU	Russian Federation		
CZ	Czech Republic	LI	Liechtenstein	SD	Sudan		
DE	Germany	LK	Sri Lanka	SE	Sweden		
DK	Denmark	LR	Liberia	SG	Singapore		
EE	Estonia						

**SYSTEM FOR ENHANCING CARDIAC SIGNAL SENSING
BY CARDIAC PACEMAKERS THROUGH GENETIC TREATMENT**

FIELD OF THE INVENTION

The present invention relates to systems for genetically enhancing cardiac signals for use by cardiac pacemakers and, more particularly, for enhancing the signal to noise ratio of atrial P-waves for improved pacemaker sensing.

BACKGROUND OF THE INVENTION

The cardiac pacemaker is a widely used device for treating various cardiac disorders, e.g., sick sinus syndrome, "brady-tachy syndrome" and heart block. The basic function of the pacemaker is to deliver stimulus pulses to one or more of the patient's heart chambers, as and when needed, to initiate cardiac depolarizations and thus maintain a desired heart rate, or to affect improvements in cardiac output for patients in heart failure. In addition to delivering stimulus pulses, another important feature is the sensing of a patient's heartbeat signals, when they occur spontaneously, for purposes of controlling the stimulus pulse delivery. Thus, the demand pacemaker inhibits delivery of a stimulus pulse and resets the pulse generator in the event of sensing a timely spontaneous beat, i.e., a P-wave which is an atrial depolarization, or a QRS, or just R-wave, which is a ventricular depolarization. For

- 2 -

example, an AAI mode pacemaker both paces and senses in just the atrium, and inhibits delivery of a pace pulse if a timely P-wave is sensed. The inhibit operation necessarily depends upon reliably sensing spontaneous P-waves. In a dual
5 chamber pacemaker, both the P-wave and R-wave are sensed. As examples of dual chamber pacemakers, see U.S. Patents 4,920,965; 4,539,991; and 4,554,921, incorporated herein by reference. A particular purpose of the dual chamber
pacemaker may be to treat a block condition, where the
10 patient's natural pacemaker is operating normally, causing timely atrial contractions, but the depolarization signal is not efficiently propagated to the ventricle so as to cause a following ventricular contraction. In such a situation, the dual chamber pacemaker is designed to sense the P-wave, and
15 deliver a synchronized ventricular stimulus pulse, i.e., a pulse which stimulates the ventricle after a timed AV delay which approximates the AV delay of a healthy heart. It is seen that reliable sensing of the P-wave is vital to this type of dual chamber pacing.

20 In yet another type of pacemaker operation, the pacemaker operates in what is referred to a VDD mode, meaning that it paces only in the ventricle, but senses both P-waves and R-waves, i.e., has single chamber pacing but dual chamber sensing. The advantage of this mode is that
25 only one lead need be positioned in the patient's heart, since no pacing pulses are delivered to the atrium. The VDD lead has the normal electrode or electrode pair at its distal end, for positioning in the ventricle; and it has a "floating" electrode (or electrode pair) proximal to the tip
30 and positioned so that it is located in the atrium, for sensing the P-wave. See, for example, U.S. Patent No. 5,127,694. However, since such a floating electrode is not necessarily embedded into or positioned adjacent the myocardium, the sensed P-wave is not as strong as for the
35 case where a separate atrial lead is used, and consequently, the reliability of sensing the P-wave is even less.

- 3 -

Atrial sensing is additionally considered to be a significant problem because of the low P-wave amplitudes commonly available and the presence of relatively large far field QRS and other "noise" signals. It is commonly
5 accepted that atrial P-wave amplitudes are relatively low compared to ventricular R-waves because of the differences in muscle mass near the electrodes. That is, ventricular R-waves are large because there is a large volume of myocardium around the electrode, whereas the atrial signal
10 is small because the underlying tissue is relatively thin. Thus, for any pacing system which senses the P wave, such as an AAI pacemaker or any dual sense mode pacemaker, reliably sensing P-waves is a major problem for which improvement has long been sought.

15 With regard to the source of the P-wave, it is noted that it is not the muscle itself that is sensed, but the electric potentials resulting from the depolarization of several myocardial cells, i.e., a net positive ion flow into myocardial cells through specialized membrane proteins
20 called voltage-gated ion channels, such as the sodium channels. More muscle mass means there are more membrane channels in the area adjacent to the electrodes. However, the muscle mass adjacent to the atrial electrode cannot be increased. But the P-wave could be enhanced if the number
25 of conducting membrane channels within the adjacent muscle mass can be increased. Sodium channels are transmembrane proteins responsible for the rapid transport of Na⁺ ions across cell membranes underlying the depolarization of the action potential in many types of cells. In particular,
30 cardiac fast sodium channels are responsible for the fast upstroke or phase 0 of the action potential in myocardial cells. Fozzard, et al., *Circ. Res.*, 1985, 56, 475-485. Recently, a human cardiac voltage-dependent sodium channel, hH1, has been cloned, sequenced, and functionally expressed.
35 Gellens, et al., *Proc. Natl. Acad. Sci. USA*, 1992, 89, 554-558.

- 4 -

Gene therapy has also recently emerged as a powerful approach to treating a variety of mammalian diseases. Direct transfer of genetic material into myocardial tissue *in vivo* has recently been demonstrated to
5 be an effective method of expressing a desired protein. For example, direct myocardial transfection of plasmid DNA by direct injection into the heart of rabbits and pigs (Gal, et al., *Lab. Invest.*, 1993, 68, 18-25), as well as of rats (Acsadi, et al., *The New Biol.*, 1991, 3, 71-81), has been
10 shown to result in expression of particular reporter gene products. In addition, direct *in vivo* gene transfer into myocardial cells has also been accomplished by directly injecting adenoviral vectors into the myocardium. French, et al., *Circulation*, 1994, 90, 2415-2424, and PCT
15 Publication WO 94/11506.

Pursuant to the above, this invention provides a system for enhancing the cardiac pacemaker atrial and/or ventricular sensing function, i.e., enhancing the signal to noise ratio of cardiac signals, and in particular the sensed
20 P-wave, through concurrent genetic treatment whereby the number of ion channels responsible for depolarization of the atrial or ventricular myocardial cells is increased. Applicants' invention is directed to delivery systems for introducing ion channel protein genetic material into
25 myocardial cells adjacent to or closest to the position of the atrial or ventricular electrode. In any particular application, the genetic material is placed so as to provide maximum benefit for sensing P-waves, or other cardiac signals, with the pacing lead used, i.e., for an AAI pacing
30 system, a lead which is fixated against the atrial wall.

SUMMARY OF THE INVENTION

In accordance with the above, a primary purpose of Applicants' claimed invention is to provide delivery systems for enhancing cardiac pacemaker signal sensing. In a
35 particular embodiment, the claimed invention provides delivery systems for enhancing cardiac pacemaker P-wave

- 5 -

sensing. Upon identifying a patient in which the signal to noise ratio for atrial or ventricular sensing is problematic, ion channel protein genetic material is selected such that expression of a selected ion channel protein in cells adjacent to the position of the atrial or ventricle electrode corrects or improves the signal to noise ratio for cardiac signal sensing. Preferably, expression of a selected ion channel protein can improve or correct the signal to noise ratio for cardiac signal sensing in either or both the ventricles and atria of all persons with pacemakers, especially those persons which have been diagnosed with a low signal to noise ratio for P-wave sensing. Improvement or correction of P-wave sensing can be manifested by an increase in the amplitude of the P-wave, or other characteristic of the cardiac signal, thus resulting in an increase of the signal to noise ratio of the signal sensed in the pacemaker atrial sensing channel. Delivery of the ion channel protein genetic material can be accomplished by adaptation of available pacing leads, such as, for example, AAI or DDD leads, as well as by specific modification of leads and catheters. Delivery of the genetic material may be affected by a pump or may be passive.

The ion channel protein genetic material used in the system and method of this invention comprises recombinant nucleic acid molecules comprising a nucleic acid molecule encoding the ion channel protein inserted into a delivery vehicle, such as, for example, plasmids or adenoviral vectors, and the appropriate regulatory elements. Alternatively, the ion channel protein genetic material comprises the ion channel protein itself. Expression of the desired ion channel protein from recombinant nucleic acid molecules is controlled by promoters, preferably cardiac tissue-specific promoter-enhancers, operably linked to the nucleic acid molecule encoding the ion channel protein. The conduction protein is preferably a sodium ion channel protein, such as, for example, the voltage-dependent sodium

- 6 -

channel hH1, which is used to correct or improve the signal to noise ratio of cardiac signals, and in particular, atrial P-wave sensing. The ion channel protein genetic material is delivered to specific sites adjacent to the atrial or
5 ventricular electrode within the heart by perfusion or injection of a therapeutically effective amount, which is that amount which corrects or improves the signal to noise ratio of the cardiac signal of the myocardial cells adjacent to the electrode. The therapeutically effective amount can
10 be delivered to the specific site in the heart in a single dose or multiple doses, as desired.

The present invention provides a delivery system for delivering a therapeutically effective amount of a predetermined ion channel protein genetic material to an
15 identified cardiac location adjacent the atrial or ventricular electrode, the genetic material being selected for amplifying the particular cardiac signal, such as, for example, the P-wave, from cardiac cells to which it is delivered, thus improving or correcting the cardiac signal
20 to noise ratio received by the sensing electrode. The delivery system includes the selected genetic material contained in a reservoir, and a catheter or electrode subsystem for delivering the genetic material from the reservoir to the identified cardiac location so as to
25 contact a plurality of cells in the proximity of the sensing electrode.

The delivery system may utilize an external reservoir for providing the genetic material, or alternately may utilize an implantable reservoir. In either embodiment,
30 a controllable pump mechanism may be provided for transferring therapeutic doses of the genetic material from the reservoir, through a catheter or electrode, and to the selected cardiac location. The pump may be a mini or micro pump located within the delivery system. Alternatively,
35 rather than using a pump mechanism, the ion channel protein genetic material can be passively delivered to the appropriate location adjacent the appropriate electrode.

- 7 -

The catheter subsystem may be of a type for direct introduction into the myocardium, as with a transthoracic procedure, or, more preferably, a endocardial catheter having a distal tip portion adapted for positioning and injecting the genetic material into the myocardium from within a heart chamber. In a preferred embodiment, the catheter distal tip has a normally withdrawn helical needle, which is extendable when positioned in the vicinity of the selected site so as to be screwed into the heart. The needle is hollow and connects with the catheter lumen so as to receive the pumped genetic material; it has one or more ports located so as to effectively release the genetic material for transduction into the cardiac area adjacent the sensing electrode. In the case of an electrode subsystem, an implantable electrode is used in place of the catheter subsystem, which is able to deliver drugs, such as steroids, or other bioactive agents, such as, for example, ion channel protein genetic material. Such implantable electrodes with drug dispensing capabilities are set forth in U.S. Patents 4,711,251, 5,458,631, 4,360,031, and 5,496,360, each of which are incorporated herein by reference. The delivery system can be used for one treatment and then removed, or can be implanted for subsequent treatments, in which latter case it is controllable by an external programmer type device. In another embodiment, the catheter or electrode subsystem may be combined with a pacing lead for sensing the patient's cardiac signals and for providing stimulus pulses.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 is a flow diagram presenting the primary steps involved in the practice of this invention, including selecting an appropriate genetic material, positioning delivery system against the heart wall, and expressing the genetic material in an appropriate dose into the determined location.

Figure 2 is a schematic representation of a delivery system in accordance with this invention,

- 8 -

illustrating delivery of genetic material into a patient's heart at the chosen location using a catheter subsystem.

Figure 3 is a schematic drawing of the distal portion of a catheter which can be used for injecting a solution carrying chosen genetic material into a patient's heart.

Figure 4 illustrates the distal end of a catheter, having a distal portion which encloses an osmotic pump.

Figure 5A is a schematic representation of a delivery system in accordance with this invention, having a combined catheter and pacing lead, with a separate pump; Figure 5B is another embodiment of a combined pacing lead and delivery catheter having a reservoir located at the distal end of the catheter.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

Applicants' invention provides delivery systems for correcting or improving cardiac signal sensing, especially the signal to noise ratio of the atrial P-wave, thus enhancing pacemaker sensing. A problematic signal to noise ratio for P-waves results from a naturally low amplitude P-wave generated in the atrium, noise from the ventricular QRS complex, muscle noise, noise from other sources, or a combination thereof. The signal to noise ratio is determined by routine and conventional techniques known to the skilled artisan. Once the specific problem has been identified in a particular patient, e.g., in any patient with a pacemaker or who is to receive a pacemaker, ion channel protein genetic material is selected such that expression of a selected ion channel protein corrects or improves the cardiac signal amplitude, thus improving or correcting the cardiac signal to noise ratio. The ion channel protein genetic material comprises either the ion channel protein itself or recombinant nucleic acid molecules comprising a nucleic acid molecule encoding the ion channel protein inserted into a delivery vehicle, such as, for example, plasmid, cosmid, YAC vector, viral vectors, and the

- 9 -

like, and the appropriate regulatory elements. In preferred embodiments of the present invention, the nucleic acid molecule encoding the ion channel protein is the full length coding sequence cDNA of an ion channel protein, and is

5 inserted into a plasmid or adenoviral vector, such as, for example, pGEM3 or pBR322, and Ad5, respectively. The regulatory elements are capable of directing expression in mammalian cells, specifically human cells. The regulatory elements include a promoter and a polyadenylation signal.

10 Expression of the desired ion channel protein is preferably controlled by cardiac tissue-specific promoter-enhancers, operably linked to the nucleic acid molecule encoding the ion channel protein. The ion channel protein is preferably a sodium channel protein, such as, for example, the hH1

15 voltage-regulated sodium channel, which is used to correct or improve the cardiac signal to noise ratio. The ion channel protein genetic material is preferably delivered in a pharmaceutical composition comprising, for example, the ion channel protein genetic material in a volume of

20 phosphate-buffered saline with 5% sucrose. In some embodiments, the ion channel protein genetic material is delivered with genetic material encoding the Na⁺/K⁺ pump, which is also inserted into an appropriate delivery vehicle. The ion channel protein genetic material may also be

25 delivered separately or in combination with class I and class IV antiarrhythmic drugs, which have been shown to increase sodium channel mRNA expression. The ion channel protein genetic material is delivered to specific sites within the heart, adjacent to the atrial or ventricular

30 electrode, by perfusion or injection of a therapeutically effective amount, which is that amount which corrects or improves the cardiac signal to noise ratio. Preferably, the therapeutically effective amount corrects or improves the P-wave signal to noise ratio. The therapeutically effective

35 amount can be delivered to the specific site in the heart in single or multiple doses, as desired, using the delivery systems of the invention.

- 10 -

The present invention comprises a delivery system for delivering a therapeutically effective amount of ion channel protein genetic material to a specific cardiac location, adjacent the atrial or ventricular electrode, in such a way as to enhance the amplitude of the cardiac signal, thus improving or correcting the signal to noise ratio. In a first embodiment, the delivery system basically comprises a reservoir subsystem for holding the genetic material, and a catheter subsystem in communication with the reservoir subsystem for placement of the genetic material in and around the identified cardiac location. In another embodiment, the delivery system basically comprises a reservoir subsystem for holding the genetic material, and an electrode subsystem in communication with the reservoir subsystem for placement of the genetic material in and around the identified cardiac location. As seen in the following discussion of several preferred embodiments, the reservoir subsystem and catheter subsystem or electrode subsystem may be separate, or they may be combined.

Preferably the reservoir contains up to 25 ml of a genetic material for delivery to the myocardium. In some applications, only a bolus of about 0.1-10 ml, or more preferably 1-5 ml, is delivered to the targeted areas. In other applications, such as where ion channel protein is being delivered in repeated doses, 25 ml or more may be used. Also, the genetic material may be diluted in a saline solution, such as, for example, phosphate-buffered saline (PBS), the reservoir holding the diluted solution for controlled delivery. Additionally, it is to be understood that the reservoir and associated control apparatus may be either implantable or external to the body, depending upon the circumstances, e.g., whether metered doses are to be administered to the patient over a period of time, or whether the delivery of the genetic material is essentially a one time treatment.

Referring now to Fig. 1, the primary steps involved in the practice of this invention are shown in the

- 11 -

flow diagram. The illustrated steps are performed following the initial diagnosis of a patient with a problematic P-wave signal to noise ratio, which can result from a low amplitude P-wave generated in the atrium, noise from the ventricular QRS complex, noise from other sources, or a combination thereof. Diagnosis can be accomplished, for example, by electrocardiography procedures. Preferably, the steps are performed in connection with all patients having cardiac pacemakers. As illustrated in block 30, the next step is to select the appropriate ion channel protein genetic material. This selection yields the "preselected genetic material." The ion channel protein genetic material is next prepared, as illustrated in block 31, by either inserting the nucleic acid molecules encoding the appropriate ion channel protein into a delivery vehicle with the appropriate regulatory elements, in the case of a recombinant nucleic acid molecule, or expressing the ion channel protein from an expression vector, in the case of the ion channel protein itself. As shown in block 32, the next step is to prepare and load the delivery system with a therapeutically effective amount of the ion channel protein genetic material. As illustrated in block 33, the next step comprises inserting the catheter, or other delivery subsystem, such as, for example, the electrode subsystem, into the patient's heart and positioning it against the heart wall. As shown in block 34, the next step comprises administering the therapeutically effective amount to the patient by contacting the appropriate location in the heart, adjacent to the atrial or ventricular electrode, using the delivery system described herein. An alternative method of administering the therapeutically effective amount of the ion channel protein genetic material is to directly inject the heart of the patient. The next step, shown in block 35, is to pace the patient in a standard manner, e.g., dual chamber synchronous pacing which includes sensing the patient's P-waves and delivering synchronized ventricular stimulus pulses, or AAI pacing. In accordance with this

- 12 -

step, it may be preferable to adjust the sensitivity of the atrial or ventricular sensing channel in accordance with the observed cardiac signal amplitude. The final step 36, which is optional, is to evaluate the response of the patient to the treatment by, for example, measuring the amplitude of the cardiac signal, such as, for example, the P-wave, by conventional electrocardiographic techniques, such as, for example, by telemetry from the implanted pulse generator. The sensitivity can then be adjusted accordingly.

Referring now to Fig. 2, there is shown an illustrative embodiment of a delivery system useful for certain applications of this invention, e.g., where larger amounts of genetic material alone or in solution are employed. A catheter 38, preferably a transvenous catheter, includes an elongated catheter body 40, suitably an insulative outer sheath which may be made of polyurethane, Teflon, silicone, or any other acceptable biocompatible plastic. The catheter has a standard lumen (illustrated in Fig. 3) extending therethrough for the length thereof, which communicates through to a hollow helical needle element 44, which is adapted for screwing into the patient's myocardium. The outer distal end of helical element 44 is open or porous, thus permitting genetic material in fluid form to be dispensed out of the end, as is discussed in more detail below in connection with Fig. 3. At the proximal end of the catheter, a fitting 46 is located, to which a Luer lock 48 is coupled. Luer lock 48 is coupled to the proximal end of sheath 40 and receives the lumen. A swivel mount 50 is mounted to Luer lock 48, allowing rotation of the catheter relative to Luer lock 52. Luer lock 52 in turn is coupled through control element 54 to a tube 58 which communicates with reservoir 55, suitably through flow control 57 and filter 56. Reservoir 55 holds a supply of the selected genetic material. Control elements 57 and 54 are used for adjustment of the pressure and flow rate, and may be mechanically or electronically controlled. Thus, unit 54 or 57 may be used to control either rate of delivery, or dosage

- 13 -

size, or both. Control unit 54 may be programmed to automatically release predetermined doses on a timed basis. Further, for an implanted system, control unit 54 may be activated from an external programmer as illustrated at 53.

5 Reference is made to international application published under the PCT, International Publication No. WO 95/05781, incorporated herein by reference, for a more detailed description of such a reservoir and catheter combination. It is to be understood that such a system is useful for this
10 invention primarily for applications where larger fluid amounts are to be expressed, e.g., where a diluted saline solution is used to wash or perfuse a selected area.

Referring now to Fig. 3, there is shown in expanded detail a schematic of the distal end of the
15 catheter of Fig. 2, illustrating the interconnection of the helical element 44 with the interior of the catheter. As illustrated, the helical needle 44 is provided with an internal lumen 59 which is in communication with the internal lumen 63L of the lead formed by tube 63. In this
20 embodiment, helical element 44 may also be a pacing electrode, in which case it is formed of conductive material and welded, or otherwise fastened, to tip element 61. Tip element 61 in turn is electrically connected to coil or coils 64, 65, which extend the length of the lead and are
25 connected to a pacemaker. An outer membrane 60 forms the outer wall of elongated catheter body 40, shown in Fig. 2. Further referring to Fig. 3, element 44 has an outlet 75 where the genetic material may be expressed, and holes or ports 76, 77, and 78 may also be utilized for providing
30 exits for the genetic material which is supplied through lumen 59 under a suitable pressure of zero up to about one atmosphere from reservoir 55 (shown in Fig. 2) and the control elements.

In practice, a catheter 38 of the form illustrated'
35 in Figs. 2 and 3 is advanced to the desired site for treatment, eg, adjacent the site where the sensing electrode is to be positioned. The catheter may be guided to the

- 14 -

indicated location by being passed down a steerable or guidable catheter having an accommodating lumen, for example as disclosed in U.S. Patent No. 5,030,204; or by means of a fixed configuration guide catheter such as illustrated in
5 U.S. Patent No. 5,104,393. Alternately, the catheter may be advanced to the desired location within the heart by means of a deflectable stylet, as disclosed in PCT Patent Application WO 93/04724, published March 18, 1993, or by a deflectable guide wire as disclosed in U.S. Patent No.
10 5,060,660. In yet another embodiment, the helical element 44 may be ordinarily retracted within a sheath at the time of guiding the catheter into the patient's heart, and extended for screwing into the heart by use of a stylet. Such extensible helical arrangements are well known in the
15 pacing art, and are commercially available.

It is to be understood that other forms of the reservoir subsystems and catheter subsystems are within the scope of this invention. Reservoir embodiments include, for example, drug dispensing irrigatable electrodes, such as
20 those described in U.S. Patent 4,360,031; electrically controllable, non-occluding, body implanting drug delivery system, such as those described in U.S. Patent No. 5,041,107; implantable drug infusion reservoir such as those described in U.S. Patent No. 5,176,641; medication delivery
25 devices such as those described in U.S. Patent 5,443,450; infusion pumps, such as SYNCHROMED® made by Medtronic, Inc.; and osmotic pumps, such as those made by Alza.

Referring now to Fig. 4, there is shown, by way of illustration, another embodiment of a delivery system having
30 a combined catheter and reservoir, useful for applications involving delivery of a relatively small bolus of genetic material, e.g., 1-5 ml. Fig. 4 illustrates the distal end of a catheter, having a distal portion 70 which encloses an osmotic pump. See U.S. Patent 4,711,251, assigned to
35 Medtronic, Inc., incorporated herein by reference. The pump includes an inner chamber 68 and an outer chamber 66, which chambers are separated by an impermeable membrane 67. A

- 15 -

semi-permeable outer membrane 72 forms the outer wall of chamber 66. The tubular portion 74 of the helical member connects to lumen 74L within inner chamber 68. A conductor 80, which runs the length of the catheter, extends into the inner chamber 68 and connects with extension 74E as shown at 74C to provide electrical contact through to element 44, in an application which the element 44 is used as a pacing electrode. A insulating cover 86 encompasses the conductor 80 from the point of contact with the semi-permeable outer membrane 72 distally. A seal 79 is provided at the point where the conductor passes through outer membrane 72 and inner membrane 67. An end cap 73, which may be integral with outer membrane 72 closes the chamber. Alternately, end cap 73 may be constructed to elute a predetermined medication, such as, for example, steroids. Steroids, such as dexamethasone sodium phosphate, beclamethasone, and the like, are used to control inflammatory processes.

In this arrangement, prior to inserting the catheter distal end into the patient's heart, the inner chamber 68 is charged with the genetic material which is to be dispensed into the myocardium. This may be done, for example, by simply inserting a micro needle through end cap 73, and inserting the desired bolus of genetic material into chamber 68. After the chamber 68 is filled and the catheter is implanted, body fluids will enter chamber 66 through membrane 72 to impart a pressure on the inner chamber 68 via the impermeable membrane 67. This results in a dispensing of the genetic material stored within chamber 68 through the lumen 74L of extension 74E and through the outlet 75 of the helical element 44. Although the preferred needle or element 44 is helical, additional configurations of needles or elements can also be used as known to those skilled in the art.

Still referring now to Fig. 4, there is illustrated another embodiment of a catheter tip useful for delivering a small bolus of the selected genetic material. In this embodiment, the bolus of material is stored within

- 16 -

the hollow interior of distal needle 44, i.e., the interior is the reservoir. The interior reservoir is maintained sealed by use of a soluble material which is normally solid, but which dissolves when subjected to body fluids for a period of time. An example of such material is mannitol. Plugs or globules 81-85 of mannitol are illustrated (by dashed lines) in place to block the two ends of element 44, as well as the ports 76, 77, 78. This may be combined with an osmotic pump, as described in connection with Fig. 3, where the outer chamber is filled with a saline solution which forces the genetic material out of the ports of element 44. Another alternate embodiment, not shown, is to use a stylet which inserted through to the distal end of the catheter, to push a piston which aids in expressing the genetic material into the myocardial cells. Alternatively, the piston can be driven by a micro pump. In another embodiment, the genetic material contacts the myocardial cells by passive delivery.

Referring now to Fig. 5A, there is shown, by way of illustration, another embodiment of an implantable delivery system comprising a combined pacing lead and delivery catheter, hereinafter referred to simply as a catheter. In this embodiment, the catheter 90 is combined with a pacemaker or pulse generator (not shown) and a source of genetic material such as illustrated by pump 92 which is suitably implanted near the pacemaker. The proximal end 91 of the catheter is connected to the pacemaker in the standard fashion. The genetic material is delivered through connecting tube 93 to a proximal section 88 of the catheter, communicating with lengthwise catheter lumen illustrated at 89. Alternately, the pacemaker head may contain a reservoir and micropump, for providing delivery of the genetic material directly to the lumen 89. The main length of the catheter has an outside sheath of biocompatible insulating material 96, and at least one conductor coil 95 which communicates electrically from the pacemaker to electrode 97 at the distal tip of the catheter. The catheter further

- 17 -

comprises an axially positioned polymeric cannula 94, having lumen 87, through at least a portion of the catheter length and positioned within coil 95, which provides an inner surface for the catheter lumen. The cannula terminates at
5 the distal end of the catheter, just proximal to the tip portion of electrode 97, which is illustrated as having an outer porous surface. Electrode 97 has a central opening, shown covered with the porous electrode material, through which genetic material can pass when the catheter is
10 positioned in the patient. As shown, conductor coil 95 is electrically connected to electrode 97, and connects pace pulses and sensed cardiac signals between the pacemaker and the electrode. Of course, for a bipolar embodiment, the lead/catheter 90 carries a second electrode (not shown),
15 suitably a ring electrode just proximal to electrode 97. Also, as illustrated, a fixation mechanism such as tines 98 are employed for fixing or anchoring the distal tip to the heart wall of the patient.

In one embodiment, pump 92 is suitably an osmotic
20 minipump, which pumps fluid contained within through tube 93, into catheter portion 88 and through the lumens 89, 87 to the tip electrode 97. As mentioned previously, the reservoir and pump may alternately be mounted in the pacemaker device itself. In either instance, the genetic
25 material is delivered under very minimal pressure from the reservoir through the lumen of the catheter to the electrode, where it is passed through the electrode central channel to contact myocardial cells. In yet another embodiment, the lumen portion 87 provided by the cannula is
30 utilized as the reservoir. In this embodiment, delivery may either be passive, or with the aid of a micropump (not shown). The genetic material can be preloaded into the cannula, or it can be inserted by a needle just before the catheter is introduced and positioned with the patient.

35 In another embodiment, as illustrated in Figure 5B, a chamber 99 is provided just proximal from eluting electrode 97, and serves as the reservoir of the genetic

- 18 -

material. Insulating material 96 is formed from a self-sealing material such that it may be pierced with a needle, or the like, and reseal itself, thus allowing introduction of the genetic material into the chamber prior to
5 implantation. Alternately, insulating material 96 can contain a port (not shown) through which the needle inserts the genetic material. In this embodiment, delivery of the material is without a pump, i.e., passive, the material draining slowly through the microporous portion of electrode
10 97.

The above described delivery systems can be used, for example, in methods of pacing and enhancing the detectability of sensed cardiac signals. A supply of a genetic material of the class having the property of
15 increasing the expression of ion channels in cardiac cells to which it is delivered is selected. A transvenous catheter, having proximal and distal ends and a pacing electrode at the distal end, is introduced into the patient. The distal end of the catheter is positioned against the
20 patient's heart wall and the genetic material is delivered through the catheter and out of the distal end, to the cardiac cells adjacent the pacing electrode, thereby enhancing cardiac signals produced by the cells. Normal cardiac pacing is carried out with the pacemaker and
25 connected catheter implanted in the patient.

Although a transvenous form of delivery system is preferred, it is to be understood that the invention can employ other methods and devices. For example, a small bolus of selected genetic material can be loaded into a
30 micro-syringe, e.g., a 100 μ l Hamilton syringe, and applied directly from the outside of the heart.

As used herein, the phrase "cardiac signal" refers to any cardiac signal that is detectable and includes, but is not limited to, the P-wave.

35 As used herein, the phrase "signal to noise ratio" refers to the ratio of the amplitude of the cardiac signal, such as, for example, the P-wave, to the amplitude of the

- 19 -

"noise." In addition, the signal to noise ratio can be measured for other cardiac signals as well. Sources of "noise" include, but are not limited to, the QRS complex and muscle noise. It is desirable to establish a high signal to noise ratio, i.e., a signal to noise ratio of greater than 1:1 for unipolar leads and greater than 3:1 for bipolar leads. It is even more preferred to establish a signal to noise ratio greater than 10:1.

As used herein, the phrase "ion channel protein genetic material" refers to recombinant nucleic acid molecules encoding an ion channel protein or, alternatively, an ion channel protein itself, which is used in the methods and delivery systems of the invention. For chronic treatment, or long term treatment, the ion channel protein genetic material will be in the form of recombinant nucleic acid molecules encoding the ion channel protein. In contrast, for acute treatment, or short term treatment, the ion channel protein genetic material will be in the form of the ion channel proteins themselves.

A "recombinant nucleic acid molecule", as used herein, is comprised of an isolated ion channel protein-encoding nucleotide sequence inserted into a delivery vehicle. Regulatory elements, such as the promoter and polyadenylation signal, are operably linked to the nucleotide sequence encoding the ion channel protein, whereby the protein is capable of being produced when the recombinant nucleic acid molecule is introduced into a cell.

The nucleic acid molecules encoding the ion channel proteins are prepared synthetically or, preferably, from isolated nucleic acid molecules, as described below. A nucleic acid is "isolated" when purified away from other cellular constituents, such as, for example, other cellular nucleic acids or proteins, by standard techniques known to those of ordinary skill in the art. The coding region of the nucleic acid molecule encoding the ion channel protein can encode a full length gene product or a subfragment thereof, or a novel mutated or fusion sequence. The protein

- 20 -

coding sequence can be a sequence endogenous to the target cell, or exogenous to the target cell. The promoter, with which the coding sequence is operably associated, may or may not be one that normally is associated with the coding
5 sequence.

The nucleic acid molecule encoding the ion channel protein is inserted into an appropriate delivery vehicle, such as, for example, an expression plasmid, cosmid, YAC vector, and the like. Almost any delivery vehicle can be
10 used for introducing nucleic acids into the cardiovascular system, including, for example, recombinant vectors, such as one based on adenovirus serotype 5, Ad5, as set forth in French, et al., *Circulation*, 1994, 90, 2414-2424, which is incorporated herein by reference. An additional protocol
15 for adenovirus-mediated gene transfer to cardiac cells is set forth in WO 94/11506, Johns, *J. Clin. Invest.*, 1995, 96, 1152-1158, and in Barr, et al., *Gene Ther.*, 1994, 1, 51-58, both of which are incorporated herein by reference. Other recombinant vectors include, for example, plasmid DNA
20 vectors, such as one derived from pGEM3 or pBR322, as set forth in Acsadi, et al., *The New Biol.*, 1991, 3, 71-81, and Gal, et al., *Lab. Invest.*, 1993, 68, 18-25, both of which are incorporated herein by reference, cDNA-containing liposomes, artificial viruses, nanoparticles, and the like.
25 It is also contemplated that ion channel proteins be injected directly into the myocardium.

The regulatory elements of the recombinant nucleic acid molecules of the invention are capable of directing expression in mammalian cells, specifically human cells.
30 The regulatory elements include a promoter and a polyadenylation signal. In addition, other elements, such as a Kozak region, may also be included in the recombinant nucleic acid molecule. Examples of polyadenylation signals useful to practice the present invention include, but are
35 not limited to, SV40 polyadenylation signals and LTR polyadenylation signals. In particular, the SV40 polyadenylation signal which is in pCEP4 plasmid

- 21 -

(Invitrogen, San Diego, CA), referred to as the SV40 polyadenylation signal, can be used.

The promoters useful in constructing the recombinant nucleic acid molecules of the invention may be constitutive or inducible. A constitutive promoter is expressed under all conditions of cell growth. Exemplary constitutive promoters include the promoters for the following genes: hypoxanthine phosphoribosyl transferase (HPRT), adenosine deaminase, pyruvate kinase, β -actin, human myosin, human hemoglobin, human muscle creatine, and others. In addition, many viral promoters function constitutively in eukaryotic cells, and include, but are not limited to, the early and late promoters of SV40, the Mouse Mammary Tumor Virus (MMTV) promoter, the long terminal repeats (LTRs) of Maloney leukemia virus, Human Immunodeficiency Virus (HIV), Cytomegalovirus (CMV) immediate early promoter, Epstein Barr Virus (EBV), Rous Sarcoma Virus (RSV), and other retroviruses, and the thymidine kinase promoter of herpes simplex virus. Other promoters are known to those of ordinary skill in the art.

Inducible promoters are expressed in the presence of an inducing agent. For example, the metallothionein promoter is induced to promote (increase) transcription in the presence of certain metal ions. Other inducible promoters are known to those of ordinary skill in the art.

Promoters and polyadenylation signals used must be functional within the cells of the mammal. In order to maximize protein production, regulatory sequences may be selected which are well suited for gene expression in the cardiac cells into which the recombinant nucleic acid molecule is administered. For example, the promoter is preferably a cardiac tissue-specific promoter-enhancer, such as, for example, cardiac isoform troponin C (cTNC) promoter. Parmacek, et al., *J. Biol. Chem.*, 1990, 265, 15970-15976, and Parmacek, et al., *Mol. Cell Biol.*, 1992, 12, 1967-1976. In addition, codons may be selected which are most efficiently transcribed in the cell. One having ordinary

- 22 -

skill in the art can produce recombinant nucleic acid molecules which are functional in the cardiac cells.

Genetic material can be introduced into a cell or "contacted" by a cell by, for example, transfection or
5 transduction procedures. Transfection refers to the acquisition by a cell of new genetic material by incorporation of added nucleic acid molecules. Transfection can occur by physical or chemical methods. Many transfection techniques are known to those of ordinary skill
10 in the art including: calcium phosphate DNA co-precipitation; DEAE-dextran DNA transfection; electroporation; naked plasmid adsorption, and cationic liposome-mediated transfection. Transduction refers to the process of transferring nucleic acid into a cell using a DNA
15 or RNA virus. Suitable viral vectors for use as transducing agents include, but are not limited to, retroviral vectors, adeno associated viral vectors, vaccinia viruses, and Semliki Forest virus vectors.

Treatment of cells, or contacting cells, with
20 recombinant nucleic acid molecules can take place *in vivo* or *ex vivo*. For *ex vivo* treatment, cells are isolated from an animal (preferably a human), transformed (*i.e.*, transduced or transfected *in vitro*) with a delivery vehicle containing a nucleic acid molecule encoding an ion channel protein, and
25 then administered to a recipient. Procedures for removing cells from mammals are well known to those of ordinary skill in the art. In addition to cells, tissue or the whole or parts of organs may be removed, treated *ex vivo* and then returned to the patient. Thus, cells, tissue or organs may
30 be cultured, bathed, perfused and the like under conditions for introducing the recombinant nucleic acid molecules of the invention into the desired cells.

For *in vivo* treatment, cells of an animal, preferably a mammal and most preferably a human, are
35 transformed *in vivo* with a recombinant nucleic acid molecule of the invention. The *in vivo* treatment may involve systemic intravenous treatment with a recombinant nucleic

- 23 -

acid molecule, local internal treatment with a recombinant nucleic acid molecule, such as by localized perfusion or topical treatment, and the like. When performing *in vivo* administration of the recombinant nucleic acid molecule, the preferred delivery vehicles are based on noncytopathic eukaryotic viruses in which nonessential or complementable genes have been replaced with the nucleic acid sequence of interest. Such noncytopathic viruses include retroviruses, the life cycle of which involves reverse transcription of genomic viral RNA into DNA with subsequent proviral integration into host cellular DNA. Retroviruses have recently been approved for human gene therapy trials. Most useful are those retroviruses that are replication-deficient (*i.e.*, capable of directing synthesis of the desired proteins, but incapable of manufacturing an infectious particle). Such genetically altered retroviral expression vectors have general utility for high-efficiency transduction of genes *in vivo*. Standard protocols for producing replication-deficient retroviruses (including the steps of incorporation of exogenous genetic material into a plasmid, transfection of a packaging cell line with plasmid, production of recombinant retroviruses by the packaging cell line, collection of viral particles from tissue culture media, and infection of the target cells with viral particles) are provided in Kriegler, M. "Gene Transfer and Expression, a Laboratory Manual", W.H. Freeman Co., New York (1990) and Murry, E.J. e.d. "Methods in Molecular Biology", Vol. 7, Humana Press, Inc., Clifton, New Jersey (1991).

A preferred virus for contacting cells in certain applications, such as in *in vivo* applications, is the adeno-associated virus, a double-stranded DNA virus. The adeno-associated virus can be engineered to be replication deficient and is capable of infecting a wide range of cell types and species. It further has advantages such as heat and lipid solvent stability, high transduction frequencies in cells of diverse lineages, including hemopoietic cells, and lack of superinfection inhibition thus allowing multiple

- 24 -

series of transductions. Recent reports indicate that the adeno-associated virus can also function in an extrachromosomal fashion.

In preferred embodiments of the present invention, the recombinant nucleic acid molecules comprising nucleic acid molecules encoding the ion channel proteins, or, in the alternative, the ion channel proteins, are delivered to cardiac cells adjacent the atrial or ventricular electrode, or both, using the delivery systems set forth above.

10 Alternatively, the ion channel protein genetic material is delivered to the cardiac cells by direct injection.

In preferred embodiments of the present invention, the nucleic acid molecules encoding the ion channel proteins comprise the full length coding sequence cDNA of an ion channel protein. Preferably, the ion channel proteins are sodium channel proteins; more preferably, the ion channel protein is the voltage-regulated sodium channel hH1. Such a nucleic acid molecule is described in the Gellens, et al., *Proc. Natl. Acad. Sci. USA*, 1992, 89, 554-558, and White, et al., *Mol. Pharmacol.*, 1991, 39, 604-608 references, both of which are incorporated herein by reference, which contain the full length amino acid sequence and cDNA sequence, respectively.

Introduction of the ion channel-encoding nucleic acid molecules or the ion channel proteins to cardiac cells adjacent the atrial or ventricular electrode will result in increased expression of sodium channels, producing a larger cardiac signal, such as, for example, P-wave, and thus, an improved or corrected signal to noise ratio.

30 Nucleic acid molecules comprising nucleotide sequences encoding hH1 sodium channel are isolated and purified according to the methods set forth in Gellens, et al., *Proc. Natl. Acad. Sci. USA*, 1992, 89, 554-558, and White, et al., *Mol. Pharmacol.*, 1991, 39, 604-608. The nucleic acid and protein sequences of hH1 sodium channel are set forth in SEQ ID NO:1 and SEQ ID NO:2, respectively. It is contemplated that nucleic acid molecules comprising

- 25 -

nucleotide sequences that are preferably at least 70% homologous, more preferably at least 80% homologous, and most preferably at least 90% homologous to the ion channel nucleotide sequences described in SEQ ID NO:1 can also be
5 used.

It is understood that minor modifications of nucleotide sequence or the primary amino acid sequence may result in proteins which have substantially equivalent or enhanced activity as compared to the ion channel proteins
10 exemplified herein. These modifications may be deliberate, as through site-directed mutagenesis, or may be accidental such as through mutations in hosts which produce the ion channel proteins. A "mutation" in a protein alters its primary structure (relative to the commonly occurring or
15 specifically described protein) due to changes in the nucleotide sequence of the DNA which encodes it. These mutations specifically include allelic variants. Mutational changes in the primary structure of a protein can result from deletions, additions, or substitutions. A "deletion"
20 is defined as a polypeptide in which one or more internal amino acid residues are absent as compared to the native sequence. An "addition" is defined as a polypeptide which has one or more additional internal amino acid residues as compared to the wild type protein. A "substitution" results
25 from the replacement of one or more amino acid residues by other residues. A protein "fragment" is a polypeptide consisting of a primary amino acid sequence which is identical to a portion of the primary sequence of the protein to which the polypeptide is related.

30 Preferred "substitutions" are those which are conservative, *i.e.*, wherein a residue is replaced by another of the same general type. As is well understood, naturally-occurring amino acids can be subclassified as acidic, basic, neutral and polar, or neutral and nonpolar and/or aromatic.
35 It is generally preferred that encoded peptides differing from the native form contain substituted codons for amino acids which are from the same group as that of the amino

- 26 -

acid replaced. Thus, in general, the basic amino acids Lys, Arg, and Histidine are interchangeable; the acidic amino acids Asp and Glu are interchangeable; the neutral polar amino acids Ser, Thr, Cys, Gln, and Asn are interchangeable; 5 the nonpolar aliphatic acids Gly, Ala, Val, Ile, and Leu are conservative with respect to each other (but because of size, Gly and Ala are more closely related and Val, Ile and Leu are more closely related), and the aromatic amino acids Phe, Trp, and Tyr are interchangeable.

10 While Pro is a nonpolar neutral amino acid, it represents difficulties because of its effects on conformation, and substitutions by or for Pro are not preferred, except when the same or similar conformational results can be obtained. Polar amino acids which represent 15 conservative changes include Ser, Thr, Gln, Asn; and to a lesser extent, Met. In addition, although classified in different categories, Ala, Gly, and Ser seem to be interchangeable, and Cys additionally fits into this group, or may be classified with the polar neutral amino acids. 20 Some substitutions by codons for amino acids from different classes may also be useful.

Once the nucleic acid molecules encoding the ion channel proteins are isolated and purified according to the methods described above, recombinant nucleic acid molecules 25 are prepared in which the desired ion channel nucleic acid molecule is incorporated into a delivery vehicle by methods known to those skilled in the art, as taught in, for example, Sambrook et al., *Molecular Cloning: A Laboratory Manual*, Second Ed. Cold Spring Harbor Press (1989). 30 Preferred delivery vehicles include, for example, plasmids (Acsadi, et al., *The New Biol.*, 1991, 3, 71-81, and Gal, et al., *Lab. Invest.*, 1993, 68, 18-25, both of which are incorporated herein by reference) and adenovirus (WO 94/11506, Johns, *J. Clin. Invest.*, 1995, 96, 1152-1158, and 35 in Barr, et al., *Gene Ther.*, 1994, 1, 51-58, each of which are incorporated herein by reference). The nucleic acid molecules encoding ion channel proteins, or ion channel

- 27 -

proteins produced therefrom, are delivered to the cardiac cells adjacent to the atrial electrode by the delivery systems of the present invention. Thus, such delivery systems of the present invention are used to contact the
5 cardiac cells adjacent the atrial electrode with recombinant nucleic acid molecules encoding an ion channel protein, or ion channel proteins.

Where the ion channel protein genetic material is in the form of ion channel proteins, such proteins can be
10 prepared in large quantities by using various standard expression systems known to those skilled in the art. Sambrook et al., *Molecular Cloning: A Laboratory Manual*, Second Ed. Cold Spring Harbor Press (1989), pp. 16.1-16.55, incorporated herein by reference.

15 The recombinant nucleic acid molecules or ion channel proteins are preferably delivered in a pharmaceutical composition. Such pharmaceutical compositions can include, for example, the recombinant nucleic acid molecule or protein in a volume of phosphate-
20 buffered saline with 5% sucrose. In other embodiments of the invention, the recombinant nucleic acid molecule or protein is delivered with suitable pharmaceutical carriers, such as those described in the most recent edition of *Remington's Pharmaceutical Sciences*, A. Osol, a standard
25 reference text in this field. The recombinant nucleic acid molecule or protein is delivered in a therapeutically effective amount. Such amount is determined experimentally and is that amount which either improves or corrects the P-wave signal to noise ratio by enhancing the P-wave amplitude
30 as a result of the increased expression of sodium channels in the cardiac cells adjacent the atrial or ventricular electrode. The amount of recombinant nucleic acid molecule or protein is preferably between 0.01 μ g and 100 mg, more preferably between 0.1 μ g and 10 mg, more preferably between
35 1 μ g and 1 mg, and most preferably between 10 μ g and 100 μ g. A single therapeutically effective amount is referred to as a bolus. Where adenovirus vectors are used, the amount of

- 28 -

recombinant nucleic acid molecule is preferably between 10^7 plaque forming units (pfu) and 10^{15} pfu, more preferably between 10^8 pfu and 10^{14} pfu, and most preferably between 10^9 pfu and 10^{12} pfu. A single therapeutically effective amount
5 of ion channel protein genetic material is referred to as a bolus. In some embodiments of the present invention, the delivery of the recombinant nucleic acid molecules or proteins is combined with steroid elution, such as with dexamethasone sodium phosphate, beclamethasone, and the
10 like, to control inflammatory processes.

In some embodiments of the invention, it may be preferred to administer, in addition to ion channel protein genetic material, delivery vehicle encoding the Na^+/K^+ pump. The Na^+/K^+ pump acts to discharge Na^+ ions from the myocardial
15 cells that have accumulated as a result of the introduction of the ion channel protein genetic material. This treatment can be optional, as determined by the skilled practitioner. cDNA encoding the alpha and beta subunits of the human Na^+/K^+ pump are set forth in Kawakami, et al., *J. Biochem.*, 1986,
20 100, 389-397, and Kawakami, et al., *Nuc. Acids Res.*, 1986, 14, 2833-2844, both of which are incorporated herein by reference. The nucleic acid and amino acid sequences for the alpha subunit are set forth in SEQ ID NO:5 and SEQ ID NO:6, respectively. The nucleic acid and amino acid
25 sequences for the beta subunit are set forth in SEQ ID NO:7 and SEQ ID NO:8, respectively. The delivery vehicles for the pump subunits can be constructed from cDNA libraries in the same manner as set forth for hH1, except that the forward primer 5'-ATGGGGAAGGGGTTGGACGTGAT-3' (SEQ ID NO:9)
30 and reverse primer 5'-ATAGTAGGTTTCCTTCTCCACCCA-3' (SEQ ID NO:10) for the alpha subunit, and the forward primer 5'-ATGGCCCGCGGGAAAGCCAAGGAG-3' (SEQ ID NO:11) and reverse primer 5'-GCTCTTAAGTTCAATTTTACATC-3' (SEQ ID NO:12) for the beta subunit are used. It is understood that other primers can
35 be used in addition to those set forth herein, as is well known to the skilled artisan. A therapeutically effective amount of the genetic material encoding the Na^+/K^+ pump is

- 29 -

delivered to the myocardial cells using the delivery systems described herein. The therapeutically effective amount is determined by the practitioner, and depends upon the results achieved with the ion channel protein genetic material.

5 In preferred embodiments of the invention, the recombinant nucleic acid molecules encoding the ion channel proteins is delivered with class I and/or class IV antiarrhythmic drugs, such as, for example, verapamil, mexiletine, and the like, or combinations thereof. These
10 drugs may be delivered subcutaneously, intravenously, injected in the immediate vicinity of the atrial electrode, or as determined by the skilled artisan. These drugs may be delivered by one injection, or in multiple injections. The amount of antiarrhythmic drugs depends upon the age, weight,
15 sex, and other characteristics of the patient, and is determined empirically by the skilled artisan. Class I and/or class IV antiarrhythmic drugs have been shown to enhance sodium ion channel expression in mammals. Duff, et al., *Mol. Pharmacol.*, 1992, 42, 570-574, and Taouis, et al.,
20 *J. Clin. Invest.*, 1991, 88, 375-378, both of which are incorporated herein by reference.

The following examples are meant to be exemplary of the preferred embodiments of the invention and are not meant to be limiting.

25 EXAMPLES

Example 1: Isolation and Purification of Nucleic Acid Molecule Encoding hH1

Nucleic acid molecules encoding hH1 are isolated and purified according to general methods well known to
30 those skilled in the art, and in particular, by the method set forth in Gellens, et al., *Proc. Natl. Acad. Sci. USA*, 1992, 89, 554-558, incorporated herein by reference. Briefly, a size selected and random-primed adult human cardiac cDNA library constructed in λ ZAPII (Stratagene) is
35 screened with cDNA probes corresponding to nucleotides 1-4385 and 5424-7076 derived from the rat muscle TTX-I isoform (rSkM2), as set forth in Kallen, et al., *Neuron*, 1990, 4,

- 30 -

233-242, incorporated herein by reference. Hybridizations are performed at 42°C for 18 hours in 50% formamide, 5X SSPE, 5X Denhardt's solution, 0.1% SDS/salmon sperm DNA, random primed ³²P-labeled probe. Filters are washed with 6X standard saline citrate, 0.1% SDS at 65°C. Plaque purified clones are rescued as pBluescript phagemids and sequenced as described in Kallen, et al., *Neuron*, 1990, 4, 233-242. A full-length hH1 construct is made in pBluescript by sequential ligation of S14 *EcoRI*-*Sac II* (nt +1 to +252), C75 *Sac II*-*KpnI* (nt +253 to +4377), and C92 *KpnI*-*EcoRI* (nt +4378 to +8491) fragments and the full length insert is moved into a modified pSP64T vector, as set forth in White, et al., *Mol. Pharmacol.*, 1991, 39, 604-608, incorporated herein by reference. Nucleotides -151 to -8 of the 5' untranslated region are deleted from the construct using exonuclease III and mung bean nuclease, as set forth in White, et al., *Mol. Pharmacol.*, 1991, 39, 604-608.

Alternatively, cDNA for hH1 may be prepared from fresh cardiac tissue. Briefly, total cellular RNA is isolated and purified (Chomczynsky, et al., *Anal. Biochem.*, 1987, 162, 156-159) from heart tissue, obtained from cardiac transplantation donors, or from salvaged tissue, and selected for poly(A) RNA (Sambrook et al., *Molecular Cloning: A Laboratory Manual*, Second Ed. Cold Spring Harbor Press (1989), pp. 7.26-7.29). cDNA corresponding to the hH1 sodium channel protein is prepared from the poly(A) cardiac RNA by reverse transcription using a GENEAMP™ PCR kit (Perkin Elmer Cetus, Norwalk, CT), or the like, using random hexamers according to the manufacturer's instructions. The specific hH1 nucleic acid molecules are amplified by the polymerase chain reaction (PCR), also using the GENEAMP™ PCR kit, or the like, using forward and reverse primers specific for hH1 according to the manufacturer's instructions. For example, the forward primer for cloning hH1 is preferably 5'-ATGGCAAACCTTCCTATTACCTCGG-3' (SEQ ID NO:3), and the reverse primer is 5'-CACGATGGACTCACGGTCCCTGTC-3' (SEQ ID NO:4). It is understood that additional primers can be used

- 31 -

for amplification as determined by those skilled in the art. These primers may be preceded at the 5' terminus by nucleotide sequences containing endonuclease restriction sites for easy incorporation into vectors. The specific ion channel nucleic acid molecules can also be amplified by PCR from human genomic DNA (Stratagene, San Diego, CA). After cutting the PCR products with the appropriate restriction endonuclease(s), the PCR products are purified by phenol:chloroform extractions, or using commercial purification kits, such as, for example, MAGIC™ Minipreps DNA Purification System (Promega, Madison, WI). The specific nucleotide sequence of the PCR products is determined by conventional DNA sequencing procedures, and the identity of the PCR products confirmed by comparison to the published sequences for the ion channel proteins.

Example 2: Insertion of Ion Channel cDNA into Delivery Vehicles

Preferably, ion channel cDNA is inserted into either plasmid or adenoviral vectors. Plasmid vectors include for example, pGEM3 or pBR322, as set forth in Acsadi, et al., *The New Biol.*, 1991, 3, 71-81, and Gal, et al., *Lab. Invest.*, 1993, 68, 18-25. Adenoviral vectors include for example, adenovirus serotype 5, Ad5, as set forth in French, et al., *Circulation*, 1994, 90, 2414-2424, and Johns, *J. Clin. Invest.*, 1995, 96, 1152-1158.

Preferably, the primers used to amplify the ion channel nucleic acid molecules are designed with unique endonuclease restriction sites located at the 5' terminus. In the absence of such design, polylinker arms, containing unique restriction sites, can be ligated thereto. After cutting the purified PCR products with the appropriate restriction endonuclease(s), the plasmid vector, comprising a polylinker, is also cut with the same restriction endonuclease(s), affording the ion channel nucleic acid molecule a site at which to ligate. In a similar manner, recombinant adenovirus (Gluzman, et al., in *Eukaryotic Viral Vectors*, Gluzman, ed., Cold Spring Harbor Press, 1982,

- 32 -

pp.187-192, French, et al., *Circulation*, 1994, 90, 2414-2424, and Johns, J. *Clin. Invest.*, 1995, 96, 1152-1158) containing ion channel cDNA molecules are prepared in accordance with standard techniques well known to those
5 skilled in the art.

It is contemplated that variations of the above-described invention may be constructed that are consistent with the spirit of the invention.

- 33 -

SEQUENCE LISTING

(1) GENERAL INFORMATION:

- (i) APPLICANTS: Ken Stokes
Josée Morissette
- (ii) TITLE OF INVENTION: SYSTEMS FOR ENHANCING CARDIAC SIGNAL
SENSING BY CARDIAC PACEMAKERS THROUGH
GENETIC TREATMENT
- (iii) NUMBER OF SEQUENCES: 12
- (iv) CORRESPONDENCE ADDRESS:
(A) ADDRESSEE: Woodcock Washburn Kurtz Mackiewicz & Norris LLP
(B) STREET: One Liberty Place - 46th Floor
(C) CITY: Philadelphia
(D) STATE: PA
(E) COUNTRY: U.S.A.
(F) ZIP: 19103
- (v) COMPUTER READABLE FORM:
(A) MEDIUM TYPE: Floppy disk
(B) COMPUTER: IBM PC compatible
(C) OPERATING SYSTEM: PC-DOS/MS-DOS
(D) SOFTWARE: WordPerfect 6.1
- (vi) CURRENT APPLICATION DATA:
(A) APPLICATION NUMBER: N/A
(B) FILING DATE: Herewith
(C) CLASSIFICATION:
- (viii) ATTORNEY/AGENT INFORMATION:
(A) NAME: Paul K. Legaard
(B) REGISTRATION NUMBER: 38,534
(C) REFERENCE/DOCKET NUMBER: MEDT-0082
- (ix) TELECOMMUNICATION INFORMATION:
(A) TELEPHONE: (215) 568-3100
(B) TELEFAX: (215) 568-3439

(2) INFORMATION FOR SEQ ID NO:1:

(i) SEQUENCE CHARACTERISTICS:

- (A) LENGTH: 6048 bases
- (B) TYPE: nucleic acid
- (C) STRANDEDNESS: double
- (D) TOPOLOGY: linear

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:1:

ATG	GCA	AAC	TTC	CTA	TTA	CCT	CGG	GGC	ACC	AGC	AGC	TTC	CGC	AGG	45
Met	Ala	Asn	Phe	Leu	Leu	Pro	Arg	Gly	Thr	Ser	Ser	Phe	Arg	Arg	
1				5					10					15	
TTC	ACA	CGG	GAG	TCC	CTG	GCA	GCC	ATC	GAG	AAG	CGC	ATG	GCG	GAG	90
Phe	Thr	Arg	Glu	Ser	Leu	Ala	Ala	Ile	Glu	Lys	Arg	Met	Ala	Glu	
			20						25					30	
AAG	CAA	GCC	CGC	GGC	TCA	ACC	ACC	TTG	CAG	GAG	AGC	CGA	GAG	GGG	135
Lys	Gln	Ala	Arg	Gly	Ser	Thr	Thr	Leu	Gln	Glu	Ser	Arg	Glu	Gly	
			35						40					45	
CTG	CCC	GAG	GAG	GAG	GCT	CCC	CGG	CCC	CAG	CTG	GAC	CTG	CAG	GCC	180
Leu	Pro	Glu	Glu	Glu	Ala	Pro	Arg	Pro	Gln	Leu	Asp	Leu	Gln	Ala	
			50						55					60	

- 34 -

TCC AAA AAG CTG CCA GAT CTC TAT GGC AAT CCA CCC CAA GAG CTC	225
Ser Lys Lys Leu Pro Asp Leu Tyr Gly Asn Pro Pro Gln Glu Leu	
65 70 75	
ATC GGA GAG CCC CTG GAG GAC CTG GAC CCC TTC TAT AGC ACC CAA	270
Ile Gly Glu Pro Leu Glu Asp Leu Asp Pro Phe Tyr Ser Thr Gln	
80 85 90	
AAG ACT TTC ATC GTA CTG AAT AAA GGC AAG ACC ATC TTC CGG TTC	315
Lys Thr Phe Ile Val Leu Asn Lys Gly Lys Thr Ile Phe Arg Phe	
95 100 105	
AGT GCC ACC AAC GCC TTG TAT GTC CTC AGT CCC TTC CAC CCA GTT	360
Ser Ala Thr Asn Ala Leu Tyr Val Leu Ser Pro Phe His Pro Val	
110 115 120	
CGG AGA GCG GCT GTG AAG ATT CTG GTT CAC TCG CTC TTC AAC ATG	405
Arg Arg Ala Ala Val Lys Ile Leu Val His Ser Leu Phe Asn Met	
125 130 135	
CTC ATC ATG TGC ACC ATC CTC ACC AAC TGC GTG TTC ATG GCC CAG	450
Leu Ile Met Cys Thr Ile Leu Thr Asn Cys Val Phe Met Ala Gln	
140 145 150	
CAC GAC CCT CCA CCC TGG ACC AAG TAT GTC GAG TAC ACC TTC ACC	495
His Asp Pro Pro Pro Trp Thr Lys Tyr Val Glu Tyr Thr Phe Thr	
155 160 165	
GCC ATT TAC ACC TTT GAG TCT CTG GTC AAG ATT CTG GCT CGA GCT	540
Ala Ile Tyr Thr Phe Glu Ser Leu Val Lys Ile Leu Ala Arg Ala	
170 175 180	
TTC TGC CTG CAC GCG TTC ACT TTC CTT CGG GAC CCA TGG AAC TGG	585
Phe Cys Leu His Ala Phe Thr Phe Leu Arg Asp Pro Trp Asn Trp	
185 190 195	
CTG GAC TTT AGT GTG ATT ATC ATG GCA TAC ACA ACT GAA TTT GTG	630
Leu Asp Phe Ser Val Ile Ile Met Ala Tyr Thr Thr Glu Phe Val	
200 205 210	
GAC CTG GGC AAT GTC TCA GCC TTA CGC ACC TTC CGA GTC CTC CGG	675
Asp Leu Gly Asn Val Ser Ala Leu Arg Thr Phe Arg Val Leu Arg	
215 220 225	
GCC CTG AAA ACT ATA TCA GTC ATT TCA GGG CTG AAG ACC ATC GTG	720
Ala Leu Lys Thr Ile Ser Val Ile Ser Gly Leu Lys Thr Ile Val	
230 235 240	
GGG GCC CTG ATC CAG TCT GTG AAG AAG CTG GCT GAT GTG ATG GTC	765
Gly Ala Leu Ile Gln Ser Val Lys Lys Leu Ala Asp Val Met Val	
245 250 255	
CTC ACA GTC TTC TGC CTC AGC GTC TTT GCC CTC ATC GGC CTG CAG	810
Leu Thr Val Phe Cys Leu Ser Val Phe Ala Leu Ile Gly Leu Gln	
260 265 270	
CTC TTC ATG GGC AAC CTA AGG CAC AAG TGT GTG CGC AAC TTC ACA	855
Leu Phe Met Gly Asn Leu Arg His Lys Cys Val Arg Asn Phe Thr	
275 280 285	
GCG CTC AAC GGC ACC AAC GGC TCC GTG GAG GCC GAC GGC TTG GTC	900
Ala Leu Asn Gly Thr Asn Gly Ser Val Glu Ala Asp Gly Leu Val	
290 295 300	
TGG GAA TCC CTG GAC CTT TAC CTC AGT GAT CCA GAA AAT TAC CTG	945
Trp Glu Ser Leu Asp Leu Tyr Leu Ser Asp Pro Glu Asn Tyr Leu	
305 310 315	

- 35 -

CTC	AAG	AAC	GGC	ACC	TCT	GAT	GTG	TTA	CTG	TGT	GGG	AAC	AGC	TCT	990
Leu	Lys	Asn	Gly	Thr	Ser	Asp	Val	Leu	Leu	Cys	Gly	Asn	Ser	Ser	
				320					325					330	
GAC	GCT	GGG	ACA	TGT	CCG	GAG	GGC	TAC	CGG	TGC	CTA	AAG	GCA	GGC	1035
Asp	Ala	Gly	Thr	Cys	Pro	Glu	Gly	Tyr	Arg	Cys	Leu	Lys	Ala	Gly	
				335					340					345	
GAG	AAC	CCC	GAC	CAC	GGC	TAC	ACC	AGC	TTC	GAT	TCC	TTT	GCC	TGG	1080
Glu	Asn	Pro	Asp	His	Gly	Tyr	Thr	Ser	Phe	Asp	Ser	Phe	Ala	Trp	
				350					355					360	
GCC	TTT	CTT	GCA	CTC	TTC	CGC	CTG	ATG	ACG	CAG	GAC	TGC	TGG	GAG	1125
Ala	Phe	Leu	Ala	Leu	Phe	Arg	Leu	Met	Thr	Gln	Asp	Cys	Trp	Glu	
				365					370					375	
CGC	CTC	TAT	CAG	CAG	ACC	CTC	AGG	TCC	GCA	GGG	AAG	ATC	TAC	ATG	1170
Arg	Leu	Tyr	Gln	Gln	Thr	Leu	Arg	Ser	Ala	Gly	Lys	Ile	Tyr	Met	
				380					385					390	
ATC	TTC	TTC	ATG	CTT	GTC	ATC	TTC	CTG	GGG	TCC	TTC	TAC	CTG	GTG	1215
Ile	Phe	Phe	Met	Leu	Val	Ile	Phe	Leu	Gly	Ser	Phe	Tyr	Leu	Val	
				395					400					405	
AAC	CTG	ATC	CTG	GCC	GTG	GTC	GCA	ATG	GCC	TAT	GAG	GAG	CAA	AAC	1260
Asn	Leu	Ile	Leu	Ala	Val	Val	Ala	Met	Ala	Tyr	Glu	Glu	Gln	Asn	
				410					415					420	
CAA	GCC	ACC	ATC	GCT	GAG	ACC	GAG	GAG	AAG	GAA	AAG	CGC	TTC	CAG	1305
Gln	Ala	Thr	Ile	Ala	Glu	Thr	Glu	Glu	Lys	Glu	Lys	Arg	Phe	Gln	
				425					430					435	
GAG	GCC	ATG	GAA	ATG	CTC	AAG	AAA	GAA	CAC	GAG	GCC	CTC	ACC	ATC	1350
Glu	Ala	Met	Glu	Met	Leu	Lys	Lys	Glu	His	Glu	Ala	Leu	Thr	Ile	
				440					445					450	
AGG	GGT	GTG	GAT	ACC	GTG	TCC	CGT	AGC	TCC	TTG	GAG	ATG	TCC	CCT	1395
Arg	Gly	Val	Asp	Thr	Val	Ser	Arg	Ser	Ser	Leu	Glu	Met	Ser	Pro	
				455					460					465	
TTG	GCC	CCA	GTA	AAC	AGC	CAT	GAG	AGA	AGA	AGC	AAG	AGG	AGA	AAA	1440
Leu	Ala	Pro	Val	Asn	Ser	His	Glu	Arg	Arg	Ser	Lys	Arg	Arg	Lys	
				470					475					480	
CGG	ATG	TCT	TCA	GGA	ACT	GAG	GAG	TGT	GGG	GAG	GAC	AGG	CTC	CCC	1485
Arg	Met	Ser	Ser	Gly	Thr	Glu	Glu	Cys	Gly	Glu	Asp	Arg	Leu	Pro	
				485					490					495	
AAG	TCT	GAC	TCA	GAA	GAT	GGT	CCC	AGA	GCA	ATG	AAT	CAT	CTC	AGC	1520
Lys	Ser	Asp	Ser	Glu	Asp	Gly	Pro	Arg	Ala	Met	Asn	His	Leu	Ser	
				500					505					510	
CTC	ACC	CGT	GGC	CTC	AGC	AGG	ACT	TCT	ATG	AAG	CCA	CGT	TCC	AGC	1565
Leu	Thr	Arg	Gly	Leu	Ser	Arg	Thr	Ser	Met	Lys	Pro	Arg	Ser	Ser	
				515					520					525	
CGC	GGG	AGC	ATT	TTC	ACC	TTT	CGC	AGG	CGA	GAC	CTG	GGT	TCT	GAA	1620
Arg	Gly	Ser	Ile	Phe	Thr	Phe	Arg	Arg	Arg	Asp	Leu	Gly	Ser	Glu	
				530					535					540	
GCA	GAT	TTT	GCA	GAT	GAT	GAA	AAC	AGC	ACA	GCG	CGG	GAG	AGC	GAG	1665
Ala	Asp	Phe	Ala	Asp	Asp	Glu	Asn	Ser	Thr	Ala	Arg	Glu	Ser	Glu	
				545					550					555	
AGC	CAC	CAC	ACA	TCA	CTG	CTG	GTG	CCC	TGG	CCC	CTG	CGC	CGG	ACC	1710
Ser	His	His	Thr	Ser	Leu	Leu	Val	Pro	Trp	Pro	Leu	Arg	Arg	Thr	
				560					565					570	

- 36 -

AGT	GCC	CAG	GGA	CAG	CCC	AGT	CCC	GGA	ACC	TCG	GCT	CCT	GGC	CAC	1755
Ser	Ala	Gln	Gly	Gln	Pro	Ser	Pro	Gly	Thr	Ser	Ala	Pro	Gly	His	
				575					580					585	
GCC	CTC	CAT	GGC	AAA	AAG	AAC	AGC	ACT	GTG	GAC	TGC	AAT	GGG	GTG	1800
Ala	Leu	His	Gly	Lys	Lys	Asn	Ser	Thr	Val	Asp	Cys	Asn	Gly	Val	
				590					595					600	
GTC	TCA	TTA	CTG	GGG	GCA	GGC	GAC	CCA	GAG	GCC	ACA	TCC	CCA	GGA	1845
Val	Ser	Leu	Leu	Gly	Ala	Gly	Asp	Pro	Glu	Ala	Thr	Ser	Pro	Gly	
				605					610					615	
AGC	CAC	CTC	CTC	CGC	CCT	GTG	ATG	CTA	GAG	CAC	CCG	CCA	GAC	ACG	1890
Ser	His	Leu	Leu	Arg	Pro	Val	Met	Leu	Glu	His	Pro	Pro	Asp	Thr	
				620					625					630	
ACC	ACG	CCA	TCG	GAG	GAG	CCA	GGC	GGC	CCC	CAG	ATG	CTG	ACC	TCC	1935
Thr	Thr	Pro	Ser	Glu	Glu	Pro	Gly	Gly	Pro	Gln	Met	Leu	Thr	Ser	
				635					640					645	
CAG	GCT	CCG	TGT	GTA	GAT	GGC	TTC	GAG	GAG	CCA	GGA	GCA	CGG	CAG	1980
Gln	Ala	Pro	Cys	Val	Asp	Gly	Phe	Glu	Glu	Pro	Gly	Ala	Arg	Gln	
				650					655					660	
CGG	GCC	CTC	AGC	GCA	GTC	AGC	GTC	CTC	ACA	AGC	GCA	CTG	GAA	GAG	2025
Arg	Ala	Leu	Ser	Ala	Val	Ser	Val	Leu	Thr	Ser	Ala	Leu	Glu	Glu	
				665					670					675	
TTA	GAG	GAG	TCT	CGC	CAC	AAG	TGT	CCA	CCA	TGC	TGG	AAC	CGT	CTC	2070
Leu	Glu	Glu	Ser	Arg	His	Lys	Cys	Pro	Pro	Cys	Trp	Asn	Arg	Leu	
				680					685					690	
GCC	CAG	CGC	TAC	CTG	ATC	TGG	GAG	TGC	TGC	CCG	CTG	TGG	ATG	TCC	2115
Ala	Gln	Arg	Tyr	Leu	Ile	Trp	Glu	Cys	Cys	Pro	Leu	Trp	Met	Ser	
				695					700					705	
ATC	AAG	CAG	GGA	GTG	AAG	TTG	GTG	GTC	ATG	GAC	CCG	TTT	ACT	GAC	2160
Ile	Lys	Gln	Gly	Val	Lys	Leu	Val	Val	Met	Asp	Pro	Phe	Thr	Asp	
				710					715					720	
CTC	ACC	ATC	ACT	ATG	TGC	ATC	GTA	CTC	AAC	ACA	CTC	TTC	ATG	GCG	2205
Leu	Thr	Ile	Thr	Met	Cys	Ile	Val	Leu	Asn	Thr	Leu	Phe	Met	Ala	
				725					730					735	
CTG	GAG	CAC	TAC	AAC	ATG	ACA	AGT	GAA	TTC	GAG	GAG	ATG	CTG	CAG	2250
Leu	Glu	His	Tyr	Asn	Met	Thr	Ser	Glu	Phe	Glu	Glu	Met	Leu	Gln	
				740					745					750	
GTC	GGA	AAC	CTG	GTC	TTC	ACA	GGG	ATT	TTC	ACA	GCA	GAG	ATG	ACC	2295
Val	Gly	Asn	Leu	Val	Phe	Thr	Gly	Ile	Phe	Thr	Ala	Glu	Met	Thr	
				755					760					765	
TTC	AAG	ATC	ATT	GCC	CTC	GAC	CCC	TAC	TAC	TAC	TTC	CAA	CAG	GGC	2340
Phe	Lys	Ile	Ile	Ala	Leu	Asp	Pro	Tyr	Tyr	Tyr	Phe	Gln	Gln	Gly	
				770					775					780	
TGG	AAC	ATC	TTC	GAC	AGC	ATC	ATC	GTC	ATC	CTT	AGC	CTC	ATG	GAG	2385
Trp	Asn	Ile	Phe	Asp	Ser	Ile	Ile	Val	Ile	Leu	Ser	Leu	Met	Glu	
				785					790					795	
CTG	GGC	CTG	TCC	CGC	ATG	AGC	AAC	TTG	TCG	GTG	CTG	CGC	TCC	TTC	2430
Leu	Gly	Leu	Ser	Arg	Met	Ser	Asn	Leu	Ser	Val	Leu	Arg	Ser	Phe	
				800					805					810	
CGC	CTG	CTG	CGG	GTC	TTC	AAG	CTG	GCC	AAA	TCA	TGG	CCC	ACC	CTG	2475
Arg	Leu	Leu	Arg	Val	Phe	Lys	Leu	Ala	Lys	Ser	Trp	Pro	Thr	Leu	
				815					820					825	

- 37 -

AAC ACA CTC ATC AAG ATC ATC GGG AAC TCA GTG GGG GCA CTG GGG	2520
Asn Thr Leu Ile Lys Ile Ile Gly Asn Ser Val Gly Ala Leu Gly	
830 835 840	
AAC CTG ACA CTG GTG CTA GCC ATC ATC GTG TTC ATC TTT GCT GTG	2565
Asn Leu Thr Leu Val Leu Ala Ile Ile Val Phe Ile Phe Ala Val	
845 850 855	
GTG GGC ATG CAG CTC TTT GGC AAG AAC TAC TCG GAG CTG AGG GAC	2610
Val Gly Met Gln Leu Phe Gly Lys Asn Tyr Ser Glu Leu Arg Asp	
860 865 870	
AGC GAC TCA GGC CTG CTG CCT CGC TGG CAC ATG ATG GAC TTC TTT	2655
Ser Asp Ser Gly Leu Leu Pro Arg Trp His Met Met Asp Phe Phe	
875 880 885	
CAT GCC TTC CTA ATC ATC TTC CGC ATC CTC TGT GGA GAG TGG ATC	2700
His Ala Phe Leu Ile Ile Phe Arg Ile Leu Cys Gly Glu Trp Ile	
890 895 900	
GAG ACC ATG TGG GAC TGC ATG GAG GTG TCG GGG CAG TCA TTA TGC	2745
Glu Thr Met Trp Asp Cys Met Glu Val Ser Gly Gln Ser Leu Cys	
905 910 915	
CTG CTG GTC TTC TTG CTT GTT ATG GTC ATT GGC AAC CTT GTG GTC	2790
Leu Leu Val Phe Leu Leu Val Met Val Ile Gly Asn Leu Val Val	
920 925 930	
CTG AAT CTC TTC CTG GCC TTG CTG CTC AGC TCC TTC AGT GCA GAC	2835
Leu Asn Leu Phe Leu Ala Leu Leu Leu Ser Ser Phe Ser Ala Asp	
935 940 945	
AAC CTC ACA GCC CCT GAT GAG GAC AGA GAG ATG AAC AAC CTC CAG	2880
Asn Leu Thr Ala Pro Asp Glu Asp Arg Glu Met Asn Asn Leu Gln	
950 955 960	
CTG GCC CTG GCC CGC ATC CAG AGG GGC CTG CGC TTT GTC AAG CGG	2925
Leu Ala Leu Ala Arg Ile Gln Arg Gly Leu Arg Phe Val Lys Arg	
965 970 975	
ACC ACC TGG GAT TTC TGC TGT GGT CTC CTG CGG CAC CGG CCT CAG	2970
Thr Thr Trp Asp Phe Cys Cys Gly Leu Leu Arg His Arg Pro Gln	
980 985 990	
AAG CCC GCA GCC CTT GCC GCC CAG GGC CAG CTG CCC AGC TGC ATT	3015
Lys Pro Ala Ala Leu Ala Ala Gln Gly Gln Leu Pro Ser Cys Ile	
995 1000 1005	
GCC ACC CCC TAC TCC CCG CCA CCC CCA GAG ACG GAG AAG GTG CCT	3060
Ala Thr Pro Tyr Ser Pro Pro Pro Pro Glu Thr Glu Lys Val Pro	
1010 1015 1020	
CCC ACC CGC AAG GAA ACA CAG TTT GAG GAA GGC GAG CAA CCA GGC	3105
Pro Thr Arg Lys Glu Thr Gln Phe Glu Glu Gly Glu Gln Pro Gly	
1025 1030 1035	
CAG GGC ACC CCC GGG GAT CCA GAC GCC GTG TGT GTG CCC ATC GCT	3150
Gln Gly Thr Pro Gly Asp Pro Glu Pro Val Cys Val Pro Ile Ala	
1040 1045 1050	
GTG GCC GAG TCA GAC ACA GAT GAC CAA GAA GAG GAT GAG GAG AAC	3195
Val Ala Glu Ser Asp Thr Asp Asp Gln Glu Glu Asp Glu Glu Asn	
1055 1060 1065	
AGC CTG GGC ACG GAG GAG GAG TCC AGC AAG CAG CAG GAA TCC CAG	3240
Ser Leu Gly Thr Glu Glu Glu Ser Ser Lys Gln Gln Glu Ser Gln	
1070 1075 1080	

- 38 -

CCT GTG TCC GGC TGG CCC AGA GGC CCT CCG GAT TCC AGG ACC TGG	3285
Pro Val Ser Gly Trp Pro Arg Gly Pro Pro Asp Ser Arg Thr Trp	
1085 1090 1095	
AGC CAG GTG TCA GCG ACT GCC TCC TCT GAG GCC GAG GCC AGT GCA	3330
Ser Gln Val Ser Ala Thr Ala Ser Ser Glu Ala Glu Ala Ser Ala	
1100 1105 1110	
TCT CAG GCC GAC TGG CGG CAG CAG TGG AAA GCG GAA CCC CAG GCC	3375
Ser Gln Ala Asp Trp Arg Gln Gln Trp Lys Ala Glu Pro Gln Ala	
1115 1120 1125	
CCA GGG TGC GGT GAG ACC CCA GAG GAC AGT TGC TCC GAG GGC AGC	3420
Pro Gly Cys Gly Glu Thr Pro Glu Asp Ser Cys Ser Glu Gly Ser	
1130 1135 1140	
ACA GCA GAC ATG ACC AAC ACC GCT GAG CTC CTG GAG CAG ATC CCT	3465
Thr Ala Asp Met Thr Asn Thr Ala Glu Leu Leu Glu Gln Ile Pro	
1145 1150 1155	
GAC CTC GGC CAG GAT GTC AAG GAC CCA GAG GAC TGC TTC ACT GAA	3510
Asp Leu Gly Gln Asp Val Lys Asp Pro Glu Asp Cys Phe Thr Glu	
1160 1165 1170	
GGC TGT GTC CGG CGC TGT CCC TGC TGT GCG GTG GAC ACC ACA CAG	3555
Gly Cys Val Arg Arg Cys Pro Cys Cys Ala Val Asp Thr Thr Gln	
1175 1180 1185	
GCC CCA GGG AAG GTC TGG TGG CGG TTG CGC AAG ACC TGC TAC CAC	3600
Ala Pro Gly Lys Val Trp Trp Arg Leu Arg Lys Thr Cys Tyr His	
1190 1195 1200	
ATC GTG GAG CAC AGC TGG TTC GAG ACA TTC ATC ATC TTC ATG ATC	3645
Ile Val Glu His Ser Trp Phe Glu Thr Phe Ile Ile Phe Met Ile	
1205 1210 1215	
CTA CTC AGC AGT GGA GCG CTG GCC TTC GAG GAC ATC TAC CTA GAG	3690
Leu Leu Ser Ser Gly Ala Leu Ala Phe Glu Asp Ile Tyr Leu Glu	
1220 1225 1230	
GAG CGG AAG ACC ATC AAG GTT CTG CTT GAG TAT GCC GAC AAG ATG	3735
Glu Arg Lys Thr Ile Lys Val Leu Leu Glu Tyr Ala Asp Lys Met	
1235 1240 1245	
TTC ACA TAT GTC TTC GTG CTG GAG ATG CTG CTC AAG TGG GTG GCC	3780
Phe Thr Tyr Val Phe Val Leu Glu Met Leu Leu Lys Trp Val Ala	
1250 1255 1260	
TAC GGC TTC AAG AAG TAC TTC ACC AAT GCC TGG TGC TGG CTC GAC	3825
Tyr Gly Phe Lys Lys Tyr Phe Thr Asn Ala Trp Cys Trp Leu Asp	
1265 1270 1275	
TTC CTC ATC GTA GAC GTC TCT CTG GTC AGC CTG GTG GCC AAC ACC	3870
Phe Leu Ile Val Asp Val Ser Leu Val Ser Leu Val Ala Asn Thr	
1280 1285 1290	
CTG GGC TTT GCC GAG ATG GGC CCC ATC AAG TCA CTG CGG ACG CTG	3915
Leu Gly Phe Ala Glu Met Gly Pro Ile Lys Ser Leu Arg Thr Leu	
1295 1300 1305	
CGT GCA CTC CGT CCT CTG AGA GCT CTG TCA CGA TTT GAG GGC ATG	3960
Arg Ala Leu Arg Pro Leu Arg Ala Leu Ser Arg Phe Glu Gly Met	
1310 1315 1320	
AGG GTG GTG GTC AAT GCC CTG GTG GGC GCC ATC CCG TCC ATC ATG	4005
Arg Val Val Val Asn Ala Leu Val Gly Ala Ile Pro Ser Ile Met	
1325 1330 1335	

- 39 -

AAC GTC CTC CTC GTC TGC CTC ATC TTC TGG CTC ATC TTC AGC ATC	4050
Asn Val Leu Leu Val Cys Leu Ile Phe Trp Leu Ile Phe Ser Ile	
1340 1345 1350	
ATG GGC GTG AAC CTC TTT GCG GGG AAG TTT GGG AGG TGC ATC AAC	4095
Met Gly Val Asn Leu Phe Ala Gly Lys Phe Gly Arg Cys Ile Asn	
1355 1360 1365	
CAG ACA GAG GGA GAC TTG CCT TTG AAC TAC ACC ATC GTG AAC AAC	4140
Gln Thr Glu Gly Asp Leu Pro Leu Asn Tyr Thr Ile Val Asn Asn	
1370 1375 1380	
AAG AGC CAG TGT GAG TCC TTG AAC TTG ACC GGA GAA TTG TAC TGG	4185
Lys Ser Gln Cys Glu Ser Leu Asn Leu Thr Gly Glu Leu Tyr Trp	
1385 1390 1395	
ACC AAG GTG AAA GTC AAC TTT GAC AAC GTG GGG GCC GGG TAC CTG	4230
Thr Lys Val Lys Val Asn Phe Asp Asn Val Gly Ala Gly Tyr Leu	
1400 1405 1410	
GCC CTT CTG CAG GTG GCA ACA TTT AAA GGC TGG ATG GAC ATT ATG	4275
Ala Leu Leu Gln Val Ala Thr Phe Lys Gly Trp Met Asp Ile Met	
1415 1420 1425	
TAT GCA GCT GTG GAC TCC AGG GGG TAT GAA GAG CAG CCT CAG TGG	4320
Tyr Ala Ala Val Asp Ser Arg Gly Tyr Glu Glu Gln Pro Gln Trp	
1430 1435 1440	
GAA TAC AAC CTC TAC ATG TAC ATC TAT TTT GTC ATT TTC ATC ATC	4365
Glu Tyr Asn Leu Tyr Met Tyr Ile Tyr Phe Val Ile Phe Ile Ile	
1445 1450 1455	
TTT GGG TCT TTC TTC ACC CTG AAC CTC TTT ATT GGT GTC ATC ATT	4410
Phe Gly Ser Phe Phe Thr Leu Asn Leu Phe Ile Gly Val Ile Ile	
1460 1465 1470	
GAC AAC TTC AAC CAA CAG AAG AAA AAG TTA GGG GGC CAG GAC ATC	4455
Asp Asn Phe Asn Gln Gln Lys Lys Lys Leu Gly Gly Gln Asp Ile	
1475 1480 1485	
TTC ATG ACA GAG GAG CAG AAG AAG TAC TAC AAT GCC ATG AAG AAG	4500
Phe Met Thr Glu Glu Gln Lys Lys Tyr Tyr Asn Ala Met Lys Lys	
1490 1495 1500	
CTG GGC TCC AAG AAG CCC CAG AAG CCC ATC CCA CGG CCC CTG AAC	4545
Leu Gly Ser Lys Lys Pro Gln Lys Pro Ile Pro Arg Pro Leu Asn	
1505 1510 1515	
AAG TAC CAG GGC TTC ATA TTC GAC ATT GTG ACC AAG CAG GCC TTT	4590
Lys Tyr Gln Gly Phe Ile Phe Asp Ile Val Thr Lys Gln Ala Phe	
1520 1525 1530	
GAC GTC ACC ATC ATG TTT CTG ATC TGC TTG AAT ATG GTG ACC ATG	4635
Asp Val Thr Ile Met Phe Leu Ile Cys Leu Asn Met Val Thr Met	
1535 1540 1545	
ATG GTG GAG ACA GAT GAC CAA AGT CCT GAG AAA ATC AAC ATC TTG	4680
Met Val Glu Thr Asp Asp Gln Ser Pro Glu Lys Ile Asn Ile Leu	
1550 1555 1560	
GCC AAG ATC AAC CTG CTC TTT GTG GCC ATC TTC ACA GGC GAG TGT	4725
Ala Lys Ile Asn Leu Leu Phe Val Ala Ile Phe Thr Gly Glu Cys	
1565 1570 1575	
ATT GTC AAG CTG GCT GCC CTG CGC CAC TAC TAC TTC ACC AAC AGC	4770
Ile Val Lys Leu Ala Ala Leu Arg His Tyr Tyr Phe Thr Asn Ser	
1580 1585 1590	

- 40 -

TGG AAT ATC TTC	GAC TTC GTG GTT GTC	ATC CTC TCC ATC GTG	GGC	4815
Trp Asn Ile Phe	Asp Phe Val Val Val	Ile Leu Ser Ile Val	Gly	
	1595	1600	1605	
ACT GTG CTC TCG	GAC ATC ATC CAG AAG	TAC TTC TTC TCC CCG	ACG	4860
Thr Val Leu Ser	Asp Ile Ile Gln Lys	Tyr Phe Phe Ser Pro	Thr	
	1610	1615	1620	
CTC TTC CGA GTC	ATC CGC CTG GCC CGA	ATA GGC CGC ATC CTC	AGA	4905
Leu Phe Arg Val	Ile Arg Leu Ala Arg	Ile Gly Arg Ile Leu	Arg	
	1625	1630	1635	
CTG ATC CGA GGG	GCC AAG GGG ATC CGC	ACG CTG CTC TTT GCC	CTC	4950
Leu Ile Arg Gly	Ala Lys Gly Ile Arg	Thr Leu Leu Phe Ala	Leu	
	1640	1645	1650	
ATG ATG TCC CTG	CCT GCC CTC TTC AAC	ATC GGG CTG CTG CTC	TTC	4995
Met Met Ser Leu	Pro Ala Leu Phe Asn	Ile Gly Leu Leu Leu	Phe	
	1655	1660	1665	
CTC GTC ATG TTC	ATC TAC TCC ATC TTT	GGC ATG GCC AAC TTC	GCT	5040
Leu Val Met Phe	Ile Tyr Ser Ile Phe	Gly Met Ala Asn Phe	Ala	
	1670	1675	1680	
TAT GTC AAG TGG	GAG GCT GGC ATC GAC	GAC ATG TTC AAC TTC	CAG	5085
Tyr Val Lys Trp	Glu Ala Gly Ile Asp	Asp Met Phe Asn Phe	Gln	
	1685	1690	1695	
ACC TTC GCC AAC	AGC ATG CTG TGC CTC	TTC CAG ATC ACC ACG	TCG	5130
Thr Phe Ala Asn	Ser Met Leu Cys Leu	Phe Gln Ile Thr Thr	Ser	
	1700	1705	1710	
GCC GGC TGG GAT	GGC CTC CTC AGC CCC	ATC CTC AAC ACT GGG	CCG	5175
Ala Gly Trp Asp	Gly Leu Leu Ser Pro	Ile Leu Asn Thr Gly	Pro	
	1715	1720	1725	
CCC TAC TGC GAC	CCC ACT CTG CCC AAC	AGC AAT GGC TCT CGG	GGG	5220
Pro Tyr Cys Asp	Pro Thr Leu Pro Asn	Ser Asn Gly Ser Arg	Gly	
	1730	1735	1740	
GAC TGC GGG AGC	CCA GCC GTG GGC ATC	CTC TTC TTC ACC ACC	TAC	5265
Asp Cys Gly Ser	Pro Ala Val Gly Ile	Leu Phe Phe Thr Thr	Tyr	
	1745	1750	1755	
ATC ATC ATC TCC	TTC CTC ATC GTG GTC	AAC ATG TAC ATT GCC	ATC	5310
Ile Ile Ile Ser	Phe Leu Ile Val Val	Asn Met Tyr Ile Ala	Ile	
	1760	1765	1770	
ATC CTG GAG AAC	TTC AGC GTG GCC ACG	GAG GAG AGC ACC GAG	CCC	5355
Ile Leu Glu Asn	Phe Ser Val Ala Thr	Glu Glu Ser Thr Glu	Pro	
	1775	1780	1785	
CTG AGT GAG GAC	GAC TTC GAT ATG TTC	TAT GAG ATC TGG GAG	AAA	5400
Leu Ser Glu Asp	Asp Phe Asp Met Phe	Tyr Glu Ile Trp Glu	Lys	
	1790	1795	1800	
TTT GAC CCA GAG	GCC ACT CAG TTT ATT	GAG TAT TCG GTC CTG	TCT	5445
Phe Asp Pro Glu	Ala Thr Gln Phe Ile	Glu Tyr Ser Val Leu	Ser	
	1805	1810	1815	
GAC TTT GCC GAC	GCC CTG TCT GAG CCA	CTC CGT ATC GCC AAG	CCC	5490
Asp Phe Ala Asp	Ala Leu Ser Glu Pro	Leu Ile Arg Ala Lys	Pro	
	1820	1825	1830	
AAC CAG ATA AGC	CTC ATC AAC ATG GAC	CTG CCC ATG GTG AGT	GGG	5535
Asn Gln Ile Ser	Leu Ile Asn Met Asp	Leu Pro Met Val Ser	Gly	
	1835	1840	1845	

- 41 -

GAC CGC ATC CAT TGC ATG GAC ATT CTC TTT GCC TTC ACC AAA AGG	5580
Asp Arg Ile His Cys Met Asp Ile Leu Phe Ala Phe Thr Lys Arg	
1850 1855 1860	
GTC CTG GGG GAG TCT GGG GAG ATG GAC GCC CTG AAG ATC CAG ATG	5625
Val Leu Gly Glu Ser Gly Glu Met Asp Ala Leu Lys Ile Gln Met	
1865 1870 1875	
GAG GAG AAG TTC ATG GCA GCC AAC CCA TCC AAG ATC TCC TAC GAG	5670
Glu Glu Lys Phe Met Ala Ala Asn Pro Ser Lys Ile Ser Tyr Glu	
1880 1885 1890	
CCC ATC ACC ACC ACA CTC CGG CGC AAG CAC GAA GAG GTG TCG GCC	5715
Pro Ile Thr Thr Thr Leu Arg Arg Lys His Glu Glu Val Ser Ala	
1895 1900 1905	
ATG GTT ATC CAG AGA GCC TTC CGC AGG CAC CTG CTG CAA CGC TCT	5760
Met Val Ile Gln Arg Ala Phe Arg Arg His Leu Leu Gln Arg Ser	
1910 1915 1920	
TTG AAG CAT GCC TCC TTC CTC TTC CGT CAG CAG GCG GGC AGC GGC	5805
Leu Lys His Ala Ser Phe Leu Phe Arg Gln Gln Ala Gly Ser Gly	
1925 1930 1935	
CTC TCC GAA GAG GAT GCC CCT GAG CGA GAG GGC CTC ATC GCC TAC	5850
Leu Ser Glu Glu Asp Ala Pro Glu Arg Glu Gly Leu Ile Ala Tyr	
1940 1945 1950	
GTG ATG AGT GAG AAC TTC TCC CGA CCC CTT GGC CCA CCC TCC AGC	5895
Val Met Ser Glu Asn Phe Ser Arg Pro Leu Gly Pro Pro Ser Ser	
1955 1960 1965	
TCC TCC ATC TCC TCC ACT TCC TTC CCA CCC TCC TAT GAC AGT GTC	5940
Ser Ser Ile Ser Ser Thr Ser Phe Pro Pro Ser Tyr Asp Ser Val	
1970 1975 1980	
ACT AGA GCC ACC AGC GAT AAC CTC CAG GTG CGG GGG TCT GAC TAC	5985
Thr Arg Ala Thr Ser Asp Asn Leu Gln Val Arg Gly Ser Asp Tyr	
1985 1990 1995	
AGC CAC AGT GAA GAT CTC GCC GAC TTC CCC CCT TCT CCG GAC AGG	6030
Ser His Ser Glu Asp Leu Ala Asp Phe Pro Pro Ser Pro Asp Arg	
2000 2005 2010	
GAC CGT GAG TCC ATC GTG	6048
Asp Arg Glu Ser Ile Val	
2015	

(2) INFORMATION FOR SEQ ID NO:2:

(i) SEQUENCE CHARACTERISTICS:

- (A) LENGTH: 2016 amino acids
- (B) TYPE: amino acid
- (C) STRANDEDNESS: single
- (D) TOPOLOGY: unknown

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:2:

Met Ala Asn Phe Leu Leu Pro Arg Gly Thr Ser Ser Phe Arg Arg	
1 5 10 15	
Phe Thr Arg Glu Ser Leu Ala Ala Ile Glu Lys Arg Met Ala Glu	
20 25 30	
Lys Gln Ala Arg Gly Ser Thr Thr Leu Gln Glu Ser Arg Glu Gly	
35 40 45	
Leu Pro Glu Glu Glu Ala Pro Arg Pro Gln Leu Asp Leu Gln Ala	
50 55 60	

- 42 -

Ser	Lys	Lys	Leu	Pro	Asp	Leu	Tyr	Gly	Asn	Pro	Pro	Gln	Glu	Leu	65	70	75
Ile	Gly	Glu	Pro	Leu	Glu	Asp	Leu	Asp	Pro	Phe	Tyr	Ser	Thr	Gln	80	85	90
Lys	Thr	Phe	Ile	Val	Leu	Asn	Lys	Gly	Lys	Thr	Ile	Phe	Arg	Phe	95	100	105
Ser	Ala	Thr	Asn	Ala	Leu	Tyr	Val	Leu	Ser	Pro	Phe	His	Pro	Val	110	115	120
Arg	Arg	Ala	Ala	Val	Lys	Ile	Leu	Val	His	Ser	Leu	Phe	Asn	Met	125	130	135
Leu	Ile	Met	Cys	Thr	Ile	Leu	Thr	Asn	Cys	Val	Phe	Met	Ala	Gln	140	145	150
His	Asp	Pro	Pro	Pro	Trp	Thr	Lys	Tyr	Val	Glu	Tyr	Thr	Phe	Thr	155	160	165
Ala	Ile	Tyr	Thr	Phe	Glu	Ser	Leu	Val	Lys	Ile	Leu	Ala	Arg	Ala	170	175	180
Phe	Cys	Leu	His	Ala	Phe	Thr	Phe	Leu	Arg	Asp	Pro	Trp	Asn	Trp	185	190	195
Leu	Asp	Phe	Ser	Val	Ile	Ile	Met	Ala	Tyr	Thr	Thr	Glu	Phe	Val	200	205	210
Asp	Leu	Gly	Asn	Val	Ser	Ala	Leu	Arg	Thr	Phe	Arg	Val	Leu	Arg	215	220	225
Ala	Leu	Lys	Thr	Ile	Ser	Val	Ile	Ser	Gly	Leu	Lys	Thr	Ile	Val	230	235	240
Gly	Ala	Leu	Ile	Gln	Ser	Val	Lys	Lys	Leu	Ala	Asp	Val	Met	Val	245	250	255
Leu	Thr	Val	Phe	Cys	Leu	Ser	Val	Phe	Ala	Leu	Ile	Gly	Leu	Gln	260	265	270
Leu	Phe	Met	Gly	Asn	Leu	Arg	His	Lys	Cys	Val	Arg	Asn	Phe	Thr	275	280	285
Ala	Leu	Asn	Gly	Thr	Asn	Gly	Ser	Val	Glu	Ala	Asp	Gly	Leu	Val	290	295	300
Trp	Glu	Ser	Leu	Asp	Leu	Tyr	Leu	Ser	Asp	Pro	Glu	Asn	Tyr	Leu	305	310	315
Leu	Lys	Asn	Gly	Thr	Ser	Asp	Val	Leu	Leu	Cys	Gly	Asn	Ser	Ser	320	325	330
Asp	Ala	Gly	Thr	Cys	Pro	Glu	Gly	Tyr	Arg	Cys	Leu	Lys	Ala	Gly	335	340	345
Glu	Asn	Pro	Asp	His	Gly	Tyr	Thr	Ser	Phe	Asp	Ser	Phe	Ala	Trp	350	355	360
Ala	Phe	Leu	Ala	Leu	Phe	Arg	Leu	Met	Thr	Gln	Asp	Cys	Trp	Glu	365	370	375
Arg	Leu	Tyr	Gln	Gln	Thr	Leu	Arg	Ser	Ala	Gly	Lys	Ile	Tyr	Met	380	385	390

- 43 -

Ile	Phe	Phe	Met	Leu	Val	Ile	Phe	Leu	Gly	Ser	Phe	Tyr	Leu	Val	395	400	405
Asn	Leu	Ile	Leu	Ala	Val	Val	Ala	Met	Ala	Tyr	Glu	Glu	Gln	Asn	410	415	420
Gln	Ala	Thr	Ile	Ala	Glu	Thr	Glu	Glu	Lys	Glu	Lys	Arg	Phe	Gln	425	430	435
Glu	Ala	Met	Glu	Met	Leu	Lys	Lys	Glu	His	Glu	Ala	Leu	Thr	Ile	440	445	450
Arg	Gly	Val	Asp	Thr	Val	Ser	Arg	Ser	Ser	Leu	Glu	Met	Ser	Pro	455	460	465
Leu	Ala	Pro	Val	Asn	Ser	His	Glu	Arg	Arg	Ser	Lys	Arg	Arg	Lys	470	475	480
Arg	Met	Ser	Ser	Gly	Thr	Glu	Glu	Cys	Gly	Glu	Asp	Arg	Leu	Pro	485	490	495
Lys	Ser	Asp	Ser	Glu	Asp	Gly	Pro	Arg	Ala	Met	Asn	His	Leu	Ser	500	505	510
Leu	Thr	Arg	Gly	Leu	Ser	Arg	Thr	Ser	Met	Lys	Pro	Arg	Ser	Ser	515	520	525
Arg	Gly	Ser	Ile	Phe	Thr	Phe	Arg	Arg	Arg	Asp	Leu	Gly	Ser	Glu	530	535	540
Ala	Asp	Phe	Ala	Asp	Asp	Glu	Asn	Ser	Thr	Ala	Arg	Glu	Ser	Glu	545	550	555
Ser	His	His	Thr	Ser	Leu	Leu	Val	Pro	Trp	Pro	Leu	Arg	Arg	Thr	560	565	570
Ser	Ala	Gln	Gly	Gln	Pro	Ser	Pro	Gly	Thr	Ser	Ala	Pro	Gly	His	575	580	585
Ala	Leu	His	Gly	Lys	Lys	Asn	Ser	Thr	Val	Asp	Cys	Asn	Gly	Val	590	595	600
Val	Ser	Leu	Leu	Gly	Ala	Gly	Asp	Pro	Glu	Ala	Thr	Ser	Pro	Gly	605	610	615
Ser	His	Leu	Leu	Arg	Pro	Val	Met	Leu	Glu	His	Pro	Pro	Asp	Thr	620	625	630
Thr	Thr	Pro	Ser	Glu	Glu	Pro	Gly	Gly	Pro	Gln	Met	Leu	Thr	Ser	635	640	645
Gln	Ala	Pro	Cys	Val	Asp	Gly	Phe	Glu	Glu	Pro	Gly	Ala	Arg	Gln	650	655	660
Arg	Ala	Leu	Ser	Ala	Val	Ser	Val	Leu	Thr	Ser	Ala	Leu	Glu	Glu	665	670	675
Leu	Glu	Glu	Ser	Arg	His	Lys	Cys	Pro	Pro	Cys	Trp	Asn	Arg	Leu	680	685	690
Ala	Gln	Arg	Tyr	Leu	Ile	Trp	Glu	Cys	Cys	Pro	Leu	Trp	Met	Ser	695	700	705
Ile	Lys	Gln	Gly	Val	Lys	Leu	Val	Val	Met	Asp	Pro	Phe	Thr	Asp	710	715	720

- 44 -

Leu Thr Ile Thr	Met Cys Ile Val Leu	Asn Thr Leu Phe Met Ala	725	730	735
Leu Glu His Tyr	Asn Met Thr Ser Glu	Phe Glu Glu Met Leu Gln	740	745	750
Val Gly Asn Leu	Val Phe Thr Gly Ile	Phe Thr Ala Glu Met Thr	755	760	765
Phe Lys Ile Ile	Ala Leu Asp Pro Tyr	Tyr Tyr Phe Gln Gln Gly	770	775	780
Trp Asn Ile Phe	Asp Ser Ile Ile Val	Ile Leu Ser Leu Met Glu	785	790	795
Leu Gly Leu Ser	Arg Met Ser Asn Leu	Ser Val Leu Arg Ser Phe	800	805	810
Arg Leu Leu Arg	Val Phe Lys Leu Ala	Lys Ser Trp Pro Thr Leu	815	820	825
Asn Thr Leu Ile	Lys Ile Ile Gly Asn	Ser Val Gly Ala Leu Gly	830	835	840
Asn Leu Thr Leu	Val Leu Ala Ile Ile	Val Phe Ile Phe Ala Val	845	850	855
Val Gly Met Gln	Leu Phe Gly Lys Asn	Tyr Ser Glu Leu Arg Asp	860	865	870
Ser Asp Ser Gly	Leu Leu Pro Arg Trp	His Met Met Asp Phe Phe	875	880	885
His Ala Phe Leu	Ile Ile Phe Arg Ile	Leu Cys Gly Glu Trp Ile	890	895	900
Glu Thr Met Trp	Asp Cys Met Glu Val	Ser Gly Gln Ser Leu Cys	905	910	915
Leu Leu Val Phe	Leu Leu Val Met Val	Ile Gly Asn Leu Val Val	920	925	930
Leu Asn Leu Phe	Leu Ala Leu Leu Leu	Ser Ser Phe Ser Ala Asp	935	940	945
Asn Leu Thr Ala	Pro Asp Glu Asp Arg	Glu Met Asn Asn Leu Gln	950	955	960
Leu Ala Leu Ala	Arg Ile Gln Arg Gly	Leu Arg Phe Val Lys Arg	965	970	975
Thr Thr Trp Asp	Phe Cys Cys Gly Leu	Leu Arg His Arg Pro Gln	980	985	990
Lys Pro Ala Ala	Leu Ala Ala Gln Gly	Gln Leu Pro Ser Cys Ile	995	1000	1005
Ala Thr Pro Tyr	Ser Pro Pro Pro Pro	Glu Thr Glu Lys Val Pro	1010	1015	1020
Pro Thr Arg Lys	Glu Thr Gln Phe Glu	Glu Gly Glu Gln Pro Gly	1025	1030	1035
Gln Gly Thr Pro	Gly Asp Pro Glu Pro	Val Cys Val Pro Ile Ala	1040	1045	1050

- 45 -

Val	Ala	Glu	Ser	Asp	Thr	Asp	Asp	Gln	Glu	Glu	Asp	Glu	Glu	Asn	
				1055					1060					1065	
Ser	Leu	Gly	Thr	Glu	Glu	Glu	Ser	Ser	Lys	Gln	Gln	Glu	Ser	Gln	
				1070					1075					1080	
Pro	Val	Ser	Gly	Trp	Pro	Arg	Gly	Pro	Pro	Asp	Ser	Arg	Thr	Trp	
				1085					1090					1095	
Ser	Gln	Val	Ser	Ala	Thr	Ala	Ser	Ser	Glu	Ala	Glu	Ala	Ser	Ala	
				1100					1105					1110	
Ser	Gln	Ala	Asp	Trp	Arg	Gln	Gln	Trp	Lys	Ala	Glu	Pro	Gln	Ala	
				1115					1120					1125	
Pro	Gly	Cys	Gly	Glu	Thr	Pro	Glu	Asp	Ser	Cys	Ser	Glu	Gly	Ser	
				1130					1135					1140	
Thr	Ala	Asp	Met	Thr	Asn	Thr	Ala	Glu	Leu	Leu	Glu	Gln	Ile	Pro	
				1145					1150					1155	
Asp	Leu	Gly	Gln	Asp	Val	Lys	Asp	Pro	Glu	Asp	Cys	Phe	Thr	Glu	
				1160					1165					1170	
Gly	Cys	Val	Arg	Arg	Cys	Pro	Cys	Cys	Ala	Val	Asp	Thr	Thr	Gln	
				1175					1180					1185	
Ala	Pro	Gly	Lys	Val	Trp	Trp	Arg	Leu	Arg	Lys	Thr	Cys	Tyr	His	
				1190					1195					1200	
Ile	Val	Glu	His	Ser	Trp	Phe	Glu	Thr	Phe	Ile	Ile	Phe	Met	Ile	
				1205					1210					1215	
Leu	Leu	Ser	Ser	Gly	Ala	Leu	Ala	Phe	Glu	Asp	Ile	Tyr	Leu	Glu	
				1220					1225					1230	
Glu	Arg	Lys	Thr	Ile	Lys	Val	Leu	Leu	Glu	Tyr	Ala	Asp	Lys	Met	
				1235					1240					1245	
Phe	Thr	Tyr	Val	Phe	Val	Leu	Glu	Met	Leu	Leu	Lys	Trp	Val	Ala	
				1250					1255					1260	
Tyr	Gly	Phe	Lys	Lys	Tyr	Phe	Thr	Asn	Ala	Trp	Cys	Trp	Leu	Asp	
				1265					1270					1275	
Phe	Leu	Ile	Val	Asp	Val	Ser	Leu	Val	Ser	Leu	Val	Ala	Asn	Thr	
				1280					1285					1290	
Leu	Gly	Phe	Ala	Glu	Met	Gly	Pro	Ile	Lys	Ser	Leu	Arg	Thr	Leu	
				1295					1300					1305	
Arg	Ala	Leu	Arg	Pro	Leu	Arg	Ala	Leu	Ser	Arg	Phe	Glu	Gly	Met	
				1310					1315					1320	
Arg	Val	Val	Val	Asn	Ala	Leu	Val	Gly	Ala	Ile	Pro	Ser	Ile	Met	
				1325					1330					1335	
Asn	Val	Leu	Leu	Val	Cys	Leu	Ile	Phe	Trp	Leu	Ile	Phe	Ser	Ile	
				1340					1345					1350	
Met	Gly	Val	Asn	Leu	Phe	Ala	Gly	Lys	Phe	Gly	Arg	Cys	Ile	Asn	
				1355					1360					1365	
Gln	Thr	Glu	Gly	Asp	Leu	Pro	Leu	Asn	Tyr	Thr	Ile	Val	Asn	Asn	
				1370					1375					1380	

- 46 -

Lys Ser Gln Cys	Glu Ser Leu Asn Leu	Thr Gly Glu Leu Tyr	Trp
1385		1390	1395
Thr Lys Val Lys	Val Asn Phe Asp Asn	Val Gly Ala Gly Tyr	Leu
1400		1405	1410
Ala Leu Leu Gln	Val Ala Thr Phe Lys	Gly Trp Met Asp Ile	Met
1415		1420	1425
Tyr Ala Ala Val	Asp Ser Arg Gly Tyr	Glu Glu Gln Pro Gln	Trp
1430		1435	1440
Glu Tyr Asn Leu	Tyr Met Tyr Ile Tyr	Phe Val Ile Phe Ile	Ile
1445		1450	1455
Phe Gly Ser Phe	Phe Thr Leu Asn Leu	Phe Ile Gly Val Ile	Ile
1460		1465	1470
Asp Asn Phe Asn	Gln Gln Lys Lys Lys	Leu Gly Gly Gln Asp	Ile
1475		1480	1485
Phe Met Thr Glu	Glu Gln Lys Lys Tyr	Tyr Asn Ala Met Lys	Lys
1490		1495	1500
Leu Gly Ser Lys	Lys Pro Gln Lys Pro	Ile Pro Arg Pro Leu	Asn
1505		1510	1515
Lys Tyr Gln Gly	Phe Ile Phe Asp Ile	Val Thr Lys Gln Ala	Phe
1520		1525	1530
Asp Val Thr Ile	Met Phe Leu Ile Cys	Leu Asn Met Val Thr	Met
1535		1540	1545
Met Val Glu Thr	Asp Asp Gln Ser Pro	Glu Lys Ile Asn Ile	Leu
1550		1555	1560
Ala Lys Ile Asn	Leu Leu Phe Val Ala	Ile Phe Thr Gly Glu	Cys
1565		1570	1575
Ile Val Lys Leu	Ala Ala Leu Arg His	Tyr Tyr Phe Thr Asn	Ser
1580		1585	1590
Trp Asn Ile Phe	Asp Phe Val Val Val	Ile Leu Ser Ile Val	Gly
1595		1600	1605
Thr Val Leu Ser	Asp Ile Ile Gln Lys	Tyr Phe Phe Ser Pro	Thr
1610		1615	1620
Leu Phe Arg Val	Ile Arg Leu Ala Arg	Ile Gly Arg Ile Leu	Arg
1625		1630	1635
Leu Ile Arg Gly	Ala Lys Gly Ile Arg	Thr Leu Leu Phe Ala	Leu
1640		1645	1650
Met Met Ser Leu	Pro Ala Leu Phe Asn	Ile Gly Leu Leu Leu	Phe
1655		1660	1665
Leu Val Met Phe	Ile Tyr Ser Ile Phe	Gly Met Ala Asn Phe	Ala
1670		1675	1680
Tyr Val Lys Trp	Glu Ala Gly Ile Asp	Asp Met Phe Asn Phe	Gln
1685		1690	1695
Thr Phe Ala Asn	Ser Met Leu Cys Leu	Phe Gln Ile Thr Thr	Ser
1700		1705	1710

- 47 -

Ala Gly Trp Asp	Gly Leu Leu Ser Pro	Ile Leu Asn Thr Gly Pro
1715		1725
Pro Tyr Cys Asp	Pro Thr Leu Pro Asn	Ser Asn Gly Ser Arg Gly
1730		1740
Asp Cys Gly Ser	Pro Ala Val Gly Ile	Leu Phe Phe Thr Thr Tyr
1745		1755
Ile Ile Ile Ser	Phe Leu Ile Val Val	Asn Met Tyr Ile Ala Ile
1760		1770
Ile Leu Glu Asn	Phe Ser Val Ala Thr	Glu Glu Ser Thr Glu Pro
1775		1785
Leu Ser Glu Asp	Asp Phe Asp Met Phe	Tyr Glu Ile Trp Glu Lys
1790		1800
Phe Asp Pro Glu	Ala Thr Gln Phe Ile	Glu Tyr Ser Val Leu Ser
1805		1815
Asp Phe Ala Asp	Ala Leu Ser Glu Pro	Leu Ile Arg Ala Lys Pro
1820		1830
Asn Gln Ile Ser	Leu Ile Asn Met Asp	Leu Pro Met Val Ser Gly
1835		1845
Asp Arg Ile His	Cys Met Asp Ile Leu	Phe Ala Phe Thr Lys Arg
1850		1860
Val Leu Gly Glu	Ser Gly Glu Met Asp	Ala Leu Lys Ile Gln Met
1865		1875
Glu Glu Lys Phe	Met Ala Ala Asn Pro	Ser Lys Ile Ser Tyr Glu
1880		1890
Pro Ile Thr Thr	Thr Leu Arg Arg Lys	His Glu Glu Val Ser Ala
1895		1905
Met Val Ile Gln	Arg Ala Phe Arg Arg	His Leu Leu Gln Arg Ser
1910		1920
Leu Lys His Ala	Ser Phe Leu Phe Arg	Gln Gln Ala Gly Ser Gly
1925		1935
Leu Ser Glu Glu	Asp Ala Pro Glu Arg	Glu Gly Leu Ile Ala Tyr
1940		1950
Val Met Ser Glu	Asn Phe Ser Arg Pro	Leu Gly Pro Pro Ser Ser
1955		1965
Ser Ser Ile Ser	Ser Thr Ser Phe Pro	Pro Ser Tyr Asp Ser Val
1970		1980
Thr Arg Ala Thr	Ser Asp Asn Leu Gln	Val Arg Gly Ser Asp Tyr
1985		1995
Ser His Ser Glu	Asp Leu Ala Asp Phe	Pro Pro Ser Pro Asp Arg
2000		2010
Asp Arg Glu Ser	Ile Val	
2015		

- (2) INFORMATION FOR SEQ ID NO:3:
 (i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 24 bases

- 48 -

- (B) TYPE: nucleic acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: linear
 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:3:

ATGGCAAAC TCGTATTACC TCGG 24

- (2) INFORMATION FOR SEQ ID NO:4:
 (i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 24 bases
 (B) TYPE: nucleic acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: linear
 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:4:

CACGATGGAC TCACGGTCCC TGTC 24

- (2) INFORMATION FOR SEQ ID NO:5:
 (I) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 3069 bases
 (B) TYPE: nucleic acid
 (C) STRANDEDNESS: double
 (D) TOPOLOGY: linear
 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:5:

ATG GGG AAG GGG GTT GGA CGT GAT AAG TAT GAG CCT GCA GCT GTT	45
Met Gly Lys Gly Val Gly Arg Asp Lys Tyr Glu Pro Ala Ala Val	
1 5 10 15	
TCA GAA CAA GGT GAT AAA AAG GGC AAA AAG GGC AAA AAA GAC AGG	90
Ser Glu Gln Glu Asp Lys Lys Glu Lys Lys Glu Lys Lys Asp Arg	
20 25 30	
GAC ATG GAT GAA CTG AAG AAA GAA GTT TCT ATG GAT GAT CAT AAA	135
Asp Met Asp Glu Leu Lys Lys Glu Val Ser Met Asp Asp His Lys	
35 40 45	
CTT AGC CTT GAT GAA CTT CAT CGT AAA TAT GGA ACA GAC TTG AGC	180
Leu Ser Leu Asp Glu Leu His Arg Lys Tyr Gly Thr Asp Leu Ser	
50 55 60	
CGG GGA TTA ACA TCT GCT CGT GCA GCT GAG ATC CTG GCG CGA GAT	225
Arg Gly Leu Thr Ser Ala Arg Ala Ala Glu Ile Leu Ala Arg Asp	
65 70 75	
GGT CCC AAC GCC CTC ACT CCC CCT CCC ACT ACT CCT GAA TGG ATC	270
Gly Pro Asn Ala Leu Thr Pro Pro Pro Thr Thr Pro Glu Trp Ile	
80 85 90	
AAG TTT TGT CGG CAG CTC TTT GGG GGG TTC TCA ATG TTA CTG TGG	315
Lys Phe Cys Arg Gln Leu Phe Gly Gly Phe Ser Met Leu Leu Trp	
95 100 105	
ATT GGA GCG ATT CTT TGT TTC TTG GCT TAT AGC ATC CAA GCT GCT	360
Ile Gly Ala Ile Leu Cys Phe Leu Ala Tyr Ser Ile Gln Ala Ala	
110 115 120	
ACA GAA GAG GAA CCT CAA AAC GAT AAT CTG TAC CTG GGT GTG GTG	405
Thr Glu Glu Glu Pro Gln Asn Asp Asn Leu Tyr Leu Gly Val Val	
125 130 135	
CTA TCA GCC GTT GTA ATC ATA ACT GGT TGC TTC TCC TAC TAT CAA	450
Leu Ser Ala Val Val Ile Ile Thr Gly Cys Phe Ser Tyr Tyr Gln	
140 145 150	

- 49 -

GAA GCT AAA AGT TCA AAG ATC ATG GAA TCC TTC AAA AAC ATG GTC	495
Glu Ala Lys Ser Ser Lys Ile Met Glu Ser Phe Lys Asn Met Val	
155 160 165	
CCT CAG CAA GCC CTT GTG ATT CGA AAT GGT GAG AAA ATG AGC ATA	540
Pro Gln Gln Ala Leu Val Ile Arg Asn Gly Glu Lys Met Ser Ile	
170 175 180	
AAT GCG GAG GAA GTT GTG GTT GGG GAT CTG GTG GAA GTA AAA GGA	585
Asn Ala Glu Glu Val Val Val Gly Asp Lue Val Glu Val Lys Gly	
185 190 195	
GGA GAC CGA ATT CCT GCT GAC CTC AGA ATC ATA TCT GCA AAT GGC	630
Gly Asp Arg Ile Pro Ala Asp Leu Arg Ile Ile Ser Ala Asn Gly	
200 205 210	
TGC AAG GTG GAT AAC TCC TCG CTC ACT GGT GAA TCA GAA CCC CAG	675
Cys Lys Val Asp Asn Ser Ser Leu Thr Gly Glu Ser Glu Pro Gln	
215 220 225	
ACT AGG TCT CCA GAT TTC ACA AAT GAA AAC CCC CTG GAG ACG AGG	720
Thr Arg Ser Pro Asp Phe Thr Asn Glu Asn Pro Leu Glu Thr Arg	
230 235 240	
AAC ATT GCC TTC TTT TCA ACA AAT TGT GTT GAA GGC ACC GCA CGT	765
Asn Ile Ala Phe Phe Ser Thr Asn Cys Val Glu Gly Thr Ala Arg	
245 250 255	
GGT ATT GTT GTC TAC ACT GGG GAT CGC ACT GTG ATG GGA AGA ATT	810
Gly Ile Val Val Tyr Thr Gly Asp Arg Thr Val Met Gly Arg Ile	
260 265 270	
GCC ACA CTT GCT TCT GGG CTG GAA GGA GGC CAG ACC CCC ATT GCT	855
Ala Thr Leu Ala Ser Gly Leu Glu Gly Gly Gln Thr Pro Ile Ala	
275 280 285	
GCA GAA ATT GAA CAT TTT ATC CAC ATC ATC ACG GGT GTG GCT GTG	900
Ala Glu Ile Glu His Phe Ile His Ile Ile Thr Gly Val Ala Val	
290 295 300	
TTC CTG GGT GTG TCT TTC TTC ATC CTT TCT CTC ATC CTT GAG TAC	945
Phe Leu Gly Val Ser Phe Phe Ile Leu Ser Leu Ile Leu Glu Tyr	
305 310 315	
ACC TGG CTT GAG GCT GTC ATC TTC CTC ATC GGT ATC ATC GTA GCC	990
Thr Trp Leu Glu Ala Val Ile Phe Leu Ile Gly Ile Ile Val Ala	
320 325 330	
AAT GTG CCG GAA GGT TTG CTG GCC ACT GTC ACG GTC TGT CTG ACA	1035
Asn Val Pro Glu Gly Leu Leu Ala Thr Val Thr Val Cys Leu Thr	
335 340 345	
CTT ACT GCC AAA CGC ATG GCA AGG AAA AAC TGC TTA GTG AAG AAC	1080
Leu Thr Ala Lys Arg Met Ala Arg Lys Asn Cys Leu Val Lys Asn	
350 355 360	
TTA GAA GCT GTG GAG ACC TTG GGG TCC ACG TCC ACC ATC TGC TCT	1125
Leu Glu Ala Val Glu Thr Leu Gly Ser Thr Ser Thr Ile Cys Ser	
365 370 375	
GAT AAA ACT GGA ACT CTG ACT CAG AAC CGG ATG ACA GTG GCC CAC	1170
Asp Lys Thr Gly Thr Leu Thr Gln Asn Arg Met Thr Val Ala His	
380 385 390	
ATG TGG TTT GAC AAT CAA ATC CAT GAA GCT GAT ACG ACA GAG AAT	1215
Met Trp Phe Asp Asn Gln Ile His Glu Ala Asp Thr Thr Glu Asn	
395 400 405	

- 50 -

CAG	AGT	GGT	GTC	TCT	TTT	GAC	AAG	ACT	TCA	GCT	ACC	TGG	CTT	GCT	1260
Gln	Ser	Gly	Val	Ser	Phe	Asp	Lys	Thr	Ser	Ala	Thr	Trp	Leu	Ala	
				410					415					420	
CTG	TCC	AGA	ATT	GCA	GGT	CTT	TGT	AAC	AGG	GCA	GTG	TTT	CAG	GCT	1305
Leu	Ser	Arg	Ile	Ala	Gly	Leu	Cys	Asn	Arg	Ala	Val	Phe	Gln	Ala	
				425					430					435	
AAC	CAG	GAA	AAC	CTA	CCT	ATT	CTT	AAG	CGG	GCA	GTT	GCA	GGA	GAT	1350
Asn	Gln	Glu	Asn	Leu	Pro	Ile	Leu	Lys	Arg	Ala	Val	Ala	Gly	Asp	
				440					445					450	
GCC	TCT	GAG	TCA	GCA	CTC	TTA	AAG	TGC	ATA	GAG	CTG	TGC	TGT	GGT	1395
Ala	Ser	Glu	Ser	Ala	Leu	Leu	Lys	Cys	Ile	Glu	Leu	Cys	Cys	Gly	
				455					460					465	
TTC	GTG	AAG	GAG	ATG	AGA	GAA	AGA	TAC	GCC	AAA	ATC	GTC	GAG	ATA	1440
Ser	Val	Lys	Glu	Met	Arg	Glu	Arg	Tyr	Ala	Lys	Ile	Val	Glu	Ile	
				470					475					480	
CCC	TTC	AAC	TCC	ACC	AAC	AAG	TAC	CAG	TTG	TCT	ATT	CAT	AAG	AAC	1485
Pro	Phe	Asn	Ser	Thr	Asn	Lys	Tyr	Gln	Leu	Ser	Ile	His	Lys	Asn	
				485					490					495	
CCC	AAC	ACA	TCG	GAG	CCC	CAA	CAC	CTG	TTG	GTG	ATG	AAG	GGC	GCC	1520
Pro	Asn	Thr	Ser	Glu	Pro	Gln	His	Leu	Leu	Val	Met	Lys	Gly	Ala	
				500					505					510	
CCA	GAA	AGG	ATC	CTA	GAC	CGT	TGC	AGC	TCT	ATC	CTC	CTC	CAC	GGC	1565
Pro	Glu	Arg	Ile	Leu	Asp	Arg	Cys	Ser	Ser	Ile	Leu	Leu	His	Gly	
				515					520					525	
AAG	GAG	CAG	CCC	CTG	GAT	GAG	GAG	CTG	AAA	GAC	GCC	TTT	CAG	AAC	1620
Lys	Glu	Gln	Pro	Leu	Asp	Glu	Glu	Leu	Lys	Asp	Ala	Phe	Gln	Asn	
				530					535					540	
GCC	TAT	TTG	GAG	CTG	GGG	GGC	CTC	GGA	GAA	CGA	GTC	CTA	GGT	TTC	1665
Ala	Tyr	Leu	Glu	Leu	Gly	Gly	Leu	Gly	Glu	Arg	Val	Leu	Gly	Phe	
				545					550					555	
TGC	CAC	CTC	TTT	CTG	CCA	GAT	GAA	CAG	TTT	CCT	GAA	GGG	TTC	CAG	1710
Cys	His	Leu	Phe	Leu	Pro	Asp	Glu	Gln	Phe	Pro	Glu	Gly	Phe	Gln	
				560					565					570	
TTT	GAC	ACT	GAC	GAT	GTG	AAT	TTC	CCT	ATC	GAT	AAT	CTG	TGC	TTC	1755
Phe	Asp	Thr	Asp	Asp	Val	Asn	Phe	Pro	Ile	Asp	Asn	Leu	Cys	Phe	
				575					580					585	
GTT	GGG	CTC	ATC	TCC	ATG	ATT	GAC	CCT	CCA	CGG	GCG	GCC	GTT	CCT	1800
Val	Gly	Leu	Ile	Ser	Met	Ile	Asp	Pro	Pro	Arg	Ala	Ala	Val	Pro	
				590					595					600	
GAT	GCC	GTG	GGC	AAA	TGT	CGA	AGT	GCT	GGA	ATT	AAG	GTC	ATC	ATG	1845
Asp	Ala	Val	Gly	Lys	Cys	Arg	Ser	Aal	Gly	Ile	Lys	Val	Ile	Met	
				605					610					615	
GTC	ACA	GGA	GAC	CAT	CCA	ATC	ACA	GCT	AAA	GCT	ATT	GCC	AAA	GGT	1890
Val	Thr	Gly	Asp	His	Pro	Ile	Thr	Ala	Lys	Ala	Ile	Ala	Lys	Gly	
				620					625					630	
GTG	GGC	ATC	ATC	TCA	GAA	GGC	ATG	GAG	ACC	GTG	GAA	GAC	ATT	GCT	1935
Val	Gly	Ile	Ile	Ser	Glu	Gly	Asn	Glu	Thr	Val	Glu	Asp	Ile	Ala	
				635					640					645	
GCC	CGC	CTC	AAC	ATC	CCA	GTC	AGC	CAG	GTG	AAC	CCC	AGG	GAT	GCC	1980
Ala	Arg	Leu	Asn	Ile	Pro	Val	Ser	Gln	Val	Asn	Pro	Arg	Asp	Ala	
				650					655					660	

- 51 -

AAG GCC TGC GTA GTA CAC GGC AGT GAT CTA AAG GAC ATG ACC TCC	2025
Lys Ala Cys Val Val His Gly Ser Asp Leu Lys Asp Met Thr Ser	
665 670 675	
GAG CAG CTG GAT GAC ATT TTG AAG TAC CAC ACT GAG ATA GTG TTT	2070
Glu Gln Leu Asp Asp Ile Leu Lys Tyr His Thr Glu Ile Val Phe	
680 685 690	
GCC AGG ACC TCC CCT CAG CAG AAG CTC ATC ATT GTG GAA GGC TGC	2115
Ala Arg Thr Ser Pro Gln Gln Lys Leu Ile Ile Val Glu Gly Cys	
695 700 705	
CAA AGA CAG GGT GCT ATC GTG GCT GTG ACT GGT GAC GGT GTG AAT	2160
Gln Arg Gln Gly Ala Ile Val Ala Val Thr Gly Asp Gly Val Asn	
710 715 720	
GAC TCT CCA GCT TTG AAG AAA GCA GAC ATT GGG GTT GCT ATG GGG	2205
Asp Ser Pro Ala Leu Lys Lys Ala Asp Ile Gly Val Ala Met Gly	
725 730 735	
ATT GCT GGC TCA GAT GTG TCC AAG CAA GCT GCT GAC ATG ATT CTT	2250
Ile Ala Gly Ser Asp Val Ser Lys Gln Ala Ala Asp Met Ile Leu	
740 745 750	
CTG GAT GAC AAC TTT GCC TCA ATT GTG ACT GGA GTA GAG GAA GGT	2295
Leu Asp Asp Asn Phe Ala Ser Ile Val Thr Gly Val Glu Glu Gly	
755 760 765	
CGT CTG ATC TTT GAT AAC TTG AAG AAA TCC ATT GCT TAT ACC TTA	2340
Arg Leu Ile Phe Asp Asn Leu Lys Lys Ser Ile Ala Tyr Thr Leu	
770 775 780	
ACC AGT AAC ATT CCC GAG ATC ACC CCG TTC CTG ATA TTT ATT ATT	2385
Thr Ser Asn Ile Pro Glu Ile Thr Pro Phe Leu Ile Phe Ile Ile	
785 790 795	
GCA AAC ATT CCA CTA CCA CTG GGG ACT GTC ACC ATC CTC TGC ATT	2430
Ala Asn Ile Pro Leu Pro Leu Gly Thr Val Thr Ile Leu Cys Ile	
800 805 810	
GAC TTG GGC ACT GAC ATG GTT CCT GCC ATC TCC CTG GCT TAT GAG	2475
Asp Leu Gly Thr Asp Met Val Pro Ala Ile Ser Leu Ala Tyr Glu	
815 820 825	
CAG GCT GAG AGT GAC ATC ATG AAG AGA CAG CCC AGA AAT CCC AAA	2520
Gln Ala Glu Ser Asp Ile Met Lys Arg Gln Pro Arg Asn Pro Lys	
830 835 840	
ACA GAC AAA CTT GTG AAT GAG CGG CTG ATC AGC ATG GCC TAT GGG	2565
Thr Asp Lys Leu Val Asn Glu Arg Leu Ile Ser Met Ala Tyr Gly	
845 850 855	
CAG ATT GGA ATG ATC CAG GCC CTG GGA GGC TTC TTT ACT TAC TTT	2610
Gln Ile Gly Met Ile Gln Ala Leu Gly Gly Phe Phe Thr Tyr Phe	
860 865 870	
GTG ATT CTG GCT GAG AAC GGC TTC CTC CCA ATT CAC CTG TTG GGC	2655
Val Ile Leu Ala Glu Asn Gly Phe Leu Pro Ile His Leu Leu Gly	
875 880 885	
CTC CGA GTG GAC TGG GAT GAC CGC TGG ATC AAC GAT GTG GAA GAC	2700
Leu Arg Val Asp Trp Asp Asp Arg Trp Ile Asn Asp Val Glu Asp	
890 895 900	
AGC TAC GGG CAG CAG TGG ACC TAT GAG CAG AGG AAA ATC GTG GAG	2745
Ser Tyr Gly Gln Gln Trp Thr Tyr Glu Gln Arg Lys Ile Val Glu	
905 910 915	

- 52 -

TTC ACC TGC CAC ACA GCC TTC TTC GTC AGT ATC GTG GTG GTG CAG	2790
Phe Thr Cys His Thr Ala Phe Phe Val Ser Ile Val Val Val Gln	
920 925 930	
TGG GCC GAC TTG GTC ATC TGT AAG ACC AGG AGG AAT TCG GTC TTC	2835
Trp Ala Asp Leu Val Ile Cys Lys Thr Arg Arg Asn Ser Val Phe	
935 940 945	
CAG CAG GGG ATG AAG AAC AAG ATC TTG ATA TTT GGC CTC TTT GAA	2880
Gln Gln Gly Met Lys Asn Lys Ile Leu Ile Phe Gly Leu Phe Glu	
950 955 960	
GAG ACA GCC CTG GCT GCT TTC CTT TCC TAC TGC CCT GGA ATG GGT	2925
Glu Thr Ala Leu Ala Phe Leu Ser Tyr Cys Pro Gly Met Gly	
965 970 975	
GTT GCT CTT AGG ATG TAT CCC CTC AAA CCT ACC TGG TGG TTC TGT	2970
Val Ala Leu Arg Met Tyr Pro Leu Lys Pro Thr Trp Trp Phe Cys	
980 985 990	
GCC TTC CCC TAC TCT CTT CTC ATC TTC GTA TAT GAC GAA GTC AGA	3015
Ala Phe Pro Tyr Ser Leu Leu Ile Phe Val Tyr Asp Glu Val Arg	
995 1000 1005	
AAA CTC ATC ATC AGG CGA CGC CCT GGC GGC TGG GTG GAG AAG GAA	3060
Lys Leu Ile Ile Arg Arg Arg Pro Gly Gly Trp Val Glu Lys Glu	
1010 1015 1020	
ACC TAC TAT	3069
Thr Tyr Tyr	

(2) INFORMATION FOR SEQ ID NO:6:

(i) SEQUENCE CHARACTERISTICS:

- (A) LENGTH:1023 amino acids
- (B) TYPE: amino acid
- (C) STRANDEDNESS: single
- (D) TOPOLOGY: unknown

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:6:

Met Gly Lys Gly Val Gly Arg Asp Lys Tyr Glu Pro Ala Ala Val	
1 5 10 15	
Ser Glu Gln Glu Asp Lys Lys Glu Lys Lys Glu Lys Lys Asp Arg	
20 25 30	
Asp Met Asp Glu Leu Lys Lys Glu Val Ser Met Asp Asp His Lys	
35 40 45	
Leu Ser Leu Asp Glu Leu His Arg Lys Tyr Gly Thr Asp Leu Ser	
50 55 60	
Arg Gly Leu Thr Ser Ala Arg Ala Ala Glu Ile Leu Ala Arg Asp	
65 70 75	
Gly Pro Asn Ala Leu Thr Pro Pro Pro Thr Thr Pro Glu Trp Ile	
80 85 90	
Lys Phe Cys Arg Gln Leu Phe Gly Gly Phe Ser Met Leu Leu Trp	
95 100 105	
Ile Gly Ala Ile Leu Cys Phe Leu Ala Tyr Ser Ile Gln Ala Ala	
110 115 120	
Thr Glu Glu Glu Pro Gln Asn Asp Asn Leu Tyr Leu Gly Val Val	
125 130 135	

- 53 -

Leu Ser Ala Val Val Ile Ile Thr Gly Cys Phe Ser Tyr Tyr Gln
 140 145 150
 Glu Ala Lys Ser Ser Lys Ile Met Glu Ser Phe Lys Asn Met Val
 155 160 165
 Pro Gln Gln Ala Leu Val Ile Arg Asn Gly Glu Lys Met Ser Ile
 170 175 180
 Asn Ala Glu Glu Val Val Val Gly Asp Lue Val Glu Val Lys Gly
 185 190 195
 Gly Asp Arg Ile Pro Ala Asp Leu Arg Ile Ile Ser Ala Asn Gly
 200 205 210
 Cys Lys Val Asp Asn Ser Ser Leu Thr Gly Glu Ser Glu Pro Gln
 215 220 225
 Thr Arg Ser Pro Asp Phe Thr Asn Glu Asn Pro Leu Glu Thr Arg
 230 235 240
 Asn Ile Ala Phe Phe Ser Thr Asn Cys Val Glu Gly Thr Ala Arg
 245 250 255
 Gly Ile Val Val Tyr Thr Gly Asp Arg Thr Val Met Gly Arg Ile
 260 265 270
 Ala Thr Leu Ala Ser Gly Leu Glu Gly Gly Gln Thr Pro Ile Ala
 275 280 285
 Ala Glu Ile Glu His Phe Ile His Ile Ile Thr Gly Val Ala Val
 290 295 300
 Phe Leu Gly Val Ser Phe Phe Ile Leu Ser Leu Ile Leu Glu Tyr
 305 310 315
 Thr Trp Leu Glu Ala Val Ile Phe Leu Ile Gly Ile Ile Val Ala
 320 325 330
 Asn Val Pro Glu Gly Leu Leu Ala Thr Val Thr Val Cys Leu Thr
 335 340 345
 Leu Thr Ala Lys Arg Met Ala Arg Lys Asn Cys Leu Val Lys Asn
 350 355 360
 Leu Glu Ala Val Glu Thr Leu Gly Ser Thr Ser Thr Ile Cys Ser
 365 370 375
 Asp Lys Thr Gly Thr Leu Thr Gln Asn Arg Met Thr Val Ala His
 380 385 390
 Met Trp Phe Asp Asn Gln Ile His Glu Ala Asp Thr Thr Glu Asn
 395 400 405
 Gln Ser Gly Val Ser Phe Asp Lys Thr Ser Ala Thr Trp Leu Ala
 410 415 420
 Leu Ser Arg Ile Ala Gly Leu Cys Asn Arg Ala Val Phe Gln Ala
 425 430 435
 Asn Gln Glu Asn Leu Pro Ile Leu Lys Arg Ala Val Ala Gly Asp
 440 445 450
 Ala Ser Glu Ser Ala Leu Leu Lys Cys Ile Glu Leu Cys Cys Gly
 455 460 465

- 54 -

Ser Val Lys Glu	Met Arg Glu Arg Tyr	Ala Lys Ile Val Glu Ile
470	475	480
Pro Phe Asn Ser	Thr Asn Lys Tyr Gln	Leu Ser Ile His Lys Asn
485	490	495
Pro Asn Thr Ser	Glu Pro Gln His Leu	Leu Val Met Lys Gly Ala
500	505	510
Pro Glu Arg Ile	Leu Asp Arg Cys Ser	Ser Ile Leu Leu His Gly
515	520	525
Lys Glu Gln Pro	Leu Asp Glu Glu Leu	Lys Asp Ala Phe Gln Asn
530	535	540
Ala Tyr Leu Glu	Leu Gly Gly Leu Gly	Glu Arg Val Leu Gly Phe
545	550	555
Cys His Leu Phe	Leu Pro Asp Glu Gln	Phe Pro Glu Gly Phe Gln
560	565	570
Phe Asp Thr Asp	Asp Val Asn Phe Pro	Ile Asp Asn Leu Cys Phe
575	580	585
Val Gly Leu Ile	Ser Met Ile Asp Pro	Pro Arg Ala Ala Val Pro
590	595	600
Asp Ala Val Gly	Lys Cys Arg Ser Aal	Gly Ile Lys Val Ile Met
605	610	615
Val Thr Gly Asp	His Pro Ile Thr Ala	Lys Ala Ile Ala Lys Gly
620	625	630
Val Gly Ile Ile	Ser Glu Gly Asn Glu	Thr Val Glu Asp Ile Ala
635	640	645
Ala Arg Leu Asn	Ile Pro Val Ser Gln	Val Asn Pro Arg Asp Ala
650	655	660
Lys Ala Cys Val	Val His Gly Ser Asp	Leu Lys Asp Met Thr Ser
665	670	675
Glu Glm Leu Asp	Asp Ile Leu Lys Tyr	His Thr Glu Ile Val Phe
680	685	690
Ala Arg Thr Ser	Pro Gln Gln Lys Leu	Ile Ile Val Glu Gly Cys
695	700	705
Gln Arg Gln Gly	Ala Ile Val Ala Val	Thr Gly Asp Gly Val Asn
710	715	720
Asp Ser Pro Ala	Leu Lys Lys Ala Asp	Ile Gly Val Ala Met Gly
725	730	735
Ile Ala Gly Ser	Asp Val Ser Lys Gln	Ala Ala Asp Met Ile Leu
740	745	750
Leu Asp Asp Asn	Phe Ala Ser Ile Val	Thr Gly Val Glu Glu Gly
755	760	765
Arg Leu Ile Phe	Asp Asn Leu Lys Lys	Ser Ile Ala Tyr Thr Leu
770	775	780
Thr Ser Asn Ile	Pro Glu Ile Thr Pro	Phe Leu Ile Phe Ile Ile
785	790	795

- 55 -

Ala Asn Ile Pro	Leu Pro Leu Gly Thr	Val Thr Ile Leu Cys	Ile
800		805	810
Asp Leu Gly Thr	Asp Met Val Pro Ala	Ile Ser Leu Ala Tyr	Glu
815		820	825
Gln Ala Glu Ser	Asp Ile Met Lys Arg	Gln Pro Arg Asn Pro	Lys
830		835	840
Thr Asp Lys Leu	Val Asn Glu Arg Leu	Ile Ser Met Ala Tyr	Gly
845		850	855
Gln Ile Gly Met	Ile Gln Ala Leu Gly	Gly Phe Phe Thr Tyr	Phe
860		865	870
Val Ile Leu Ala	Glu Asn Gly Phe Leu	Pro Ile His Leu Leu	Gly
875		880	885
Leu Arg Val Asp	Trp Asp Asp Arg Trp	Ile Asn Asp Val Glu	Asp
890		895	900
Ser Tyr Gly Gln	Gln Trp Thr Tyr Glu	Gln Arg Lys Ile Val	Glu
905		910	915
Phe Thr Cys His	Thr Ala Phe Phe Val	Ser Ile Val Val Val	Gln
920		925	930
Trp Ala Asp Leu	Val Ile Cys Lys Thr	Arg Arg Asn Ser Val	Phe
935		940	945
Gln Gln Gly Met	Lys Asn Lys Ile Leu	Ile Phe Gly Leu Phe	Glu
950		955	960
Glu Thr Ala Leu	Ala Ala Phe Leu Ser	Tyr Cys Pro Gly Met	Gly
965		970	975
Val Ala Leu Arg	Met Tyr Pro Leu Lys	Pro Thr Trp Trp Phe	Cys
980		985	990
Ala Phe Pro Tyr	Ser Leu Leu Ile Phe	Val Tyr Asp Glu Val	Arg
995		1000	1005
Lys Leu Ile Ile	Arg Arg Arg Pro Gly	Gly Trp Val Glu Lys	Glu
1010		1015	1020
Thr Tyr Tyr			

(2) INFORMATION FOR SEQ ID NO:7:

(I) SEQUENCE CHARACTERISTICS:

- (A) LENGTH: 909 bases
- (B) TYPE: nucleic acid
- (C) STRANDEDNESS: double
- (D) TOPOLOGY: linear

(xi) SEQUENCE DESCRIPTION: SEQ ID NO:7:

ATG GCC CGC GGG AAA GCC AAG GAG GAG GGC AGC TGG AAG AAA TTC	45
Met Ala Arg Gly Lys Ala Lys Glu Glu Gly Ser Trp Lys Lys Phe	
1 5 10 15	
ATC TGG AAC TCA GAG AAG AAG GAG TTT CTG GGC AGG ACC GGT GGC	90
Ile Trp Asn Ser Glu Lys Lys Glu Phe Leu Gly Arg Thr Gly Gly	
20 25 30	
AGT TGG TTT AAG ATC CTT CTA TTC TAC GTA ATA TTT TAT GGC TGC	135
Ser Trp Phe Lys Ile Leu Leu Phe Tyr Val Ile Phe Tyr Gly Cys	
35 40 45	

- 56 -

CTG GCT GGC ATC TTC ATC GGA ACC ATC CAA GTG ATG CTG CTC ACC	180
Leu Ala Gly Ile Phe Ile Gly Thr Ile Gln Val Met Leu Leu Thr	
50 55 60	
ATC AGT GAA TTT AAG CCC ACA TAT CAG GAC CGA GTG GCC CCG CCA	225
Ile Ser Glu Phe Lys Pro Thr Tyr Gln Asp Arg Val Ala Pro Pro	
65 70 75	
GGA TTA ACA CAG ATT CCT CAG ATC CAG AAG ACT GAA ATT TCC TTT	270
Gly Leu Thr Gln Ile Pro Gln Ile Gln Lys Thr Glu Ile Ser Phe	
80 85 90	
CGT CCT AAT GAT CCC AAG AGC TAT GAG GCA TAT GTA CTG AAC ATA	315
Arg Pro Asn Asp Pro Lys Ser Tyr Glu Ala Tyr Val Leu Asn Ile	
95 100 105	
GTT AGG TTC CTG GAA AAG TAC AAA GAT TCA GCC CAG AGG GAT GAC	360
Val Arg Phe Leu Glu Lys Tyr Lys Asp Ser Ala Gln Arg Asp Asp	
110 115 120	
ATG ATT TTT GAA GAT TGT GGC GAT GTG CCC AGT GAA CCG AAA GAA	405
Met Ile Phe Glu Asp Cys Gly Asp Val Pro Ser Glu Pro Lys Glu	
125 130 135	
CGA GGA GAC TTT AAT CAT GAA CGA GGA GAG CGA AAG GTC TGC AGA	450
Arg Gly Asp Phe Asn His Glu Arg Gly Glu Arg Lys Val Cys Arg	
140 145 150	
TTC AAG CTT GAA TGG CTG GGA AAT TGC TCT GGA TTA AAT GAT GAA	495
Phy Lys Leu Glu Trp Leu Gly Asn Cys Ser Gly Leu Asn Asp Glu	
155 160 165	
ACT TAT GGC TAC AAA GAG GGC AAA CCG TGC ATT ATT ATA AAG CTC	540
Thr Tyr Gly Tyr Lys Glu Gly Lys Pro Cys Ile Ile Ile Lys Leu	
170 175 180	
AAC CGA GTT CTA GGC TTC AAA CCT AAG CCT CCC AAG AAT GAG TCC	585
Asn Arg Val Leu Gly Phe Lys Pro Lys Pro Pro Lys Asn Glu Ser	
185 190 195	
TTG GAG ACT TAC CCA GTG ATG AAG TAT AAC CCA AAT GTC CTT CCC	630
Leu Glu Thr Tyr Pro Val Met Lys Tyr Asn Pro Asn Val Leu Pro	
200 205 210	
GTT CAG TGC ACT GGC AAG CGA GAT GAA GAT AAG GAT AAA GTT GGA	675
Val Gln Cys Thr Gly Lys Arg Asp Glu Asp Lys Asp Lys Val Gly	
215 220 225	
AAT GTG GAG TAT TTT GGA CTG GGC AAC TCC CCT GGT TTT CCT CTG	720
Asn Val Glu Tyr Phe Gly Leu Gly Asn Ser Pro Gly Phe Pro Leu	
230 235 240	
CAG TAT TAT CCG TAC TAT GGC AAA CTC CTG CAG CCC AAA TAC CTG	765
Gln Tyr Tyr Pro Tyr Tyr Gly Lys Leu Leu Gln Pro Lys Tyr Leu	
245 250 255	
CAG CCC CTG CTG GCC GTA CAG TTC ACC AAT CTT ACC ATG GAC ACT	810
Gln Pro Leu Leu Ala Val Gln Phe Thr Asn Leu Thr Met Asp Thr	
260 265 270	
GAA ATT CGC ATA GAG TGT AAG GCG TAC GGT GAG AAC ATT GGG TAC	855
Glu Ile Arg Ile Glu Cys Lys Ala Tyr Gly Glu Asn Ile Gly Tyr	
275 280 285	
AGT GAG AAA GAC CGT TTT CAG GGA CGT TTT GAT GTA AAA ATT GAA	900
Ser Glu Lys Asp Arg Phe Gln Gly Arg Phe Asp Val Lys Ile Glu	
290 295 300	

- 57 -

GTT AAG AGC 909
Val Lys Ser

- (2) INFORMATION FOR SEQ ID NO:8:
 (i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH:303 amino acids
 (B) TYPE: amino acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: unknown
 (xi) SEQUENCE DESCRIPTION: SEQ ID NO:8:

Met	Ala	Arg	Gly	Lys	Ala	Lys	Glu	Glu	Gly	Ser	Trp	Lys	Lys	Phe	1	5	10	15
Ile	Trp	Asn	Ser	Glu	Lys	Lys	Glu	Phe	Leu	Gly	Arg	Thr	Gly	Gly	20	25	30	
Ser	Trp	Phe	Lys	Ile	Leu	Leu	Phe	Tyr	Val	Ile	Phe	Tyr	Gly	Cys	35	40	45	
Leu	Ala	Gly	Ile	Phe	Ile	Gly	Thr	Ile	Gln	Val	Met	Leu	Leu	Thr	50	55	60	
Ile	Ser	Glu	Phe	Lys	Pro	Thr	Tyr	Gln	Asp	Arg	Val	Ala	Pro	Pro	65	70	75	
Gly	Leu	Thr	Gln	Ile	Pro	Gln	Ile	Gln	Lys	Thr	Glu	Ile	Ser	Phe	80	85	90	
Arg	Pro	Asn	Asp	Pro	Lys	Ser	Tyr	Glu	Ala	Tyr	Val	Leu	Asn	Ile	95	100	105	
Val	Arg	Phe	Leu	Glu	Lys	Tyr	Lys	Asp	Ser	Ala	Gln	Arg	Asp	Asp	110	115	120	
Met	Ile	Phe	Glu	Asp	Cys	Gly	Asp	Val	Pro	Ser	Glu	Pro	Lys	Glu	125	130	135	
Arg	Gly	Asp	Phe	Asn	His	Glu	Arg	Gly	Glu	Arg	Lys	Val	Cys	Arg	140	145	150	
Phy	Lys	Leu	Glu	Trp	Leu	Gly	Asn	Cys	Ser	Gly	Leu	Asn	Asp	Glu	155	160	165	
Thr	Tyr	Gly	Tyr	Lys	Glu	Gly	Lys	Pro	Cys	Ile	Ile	Ile	Lys	Leu	170	175	180	
Asn	Arg	Val	Leu	Gly	Phe	Lys	Pro	Lys	Pro	Pro	Lys	Asn	Glu	Ser	185	190	195	
Leu	Glu	Thr	Tyr	Pro	Val	Met	Lys	Tyr	Asn	Pro	Asn	Val	Leu	Pro	200	205	210	
Val	Gln	Cys	Thr	Gly	Lys	Arg	Asp	Glu	Asp	Lys	Asp	Lys	Val	Gly	215	220	225	
Asn	Val	Glu	Tyr	Phe	Gly	Leu	Gly	Asn	Ser	Pro	Gly	Phe	Pro	Leu	230	235	240	
Gln	Tyr	Tyr	Pro	Tyr	Tyr	Gly	Lys	Leu	Leu	Gln	Pro	Lys	Tyr	Leu	245	250	255	
Gln	Pro	Leu	Leu	Ala	Val	Gln	Phe	Thr	Asn	Leu	Thr	Met	Asp	Thr	260	265	270	

- 58 -

Glu Ile Arg Ile Glu Cys Lys Ala Tyr Gly Glu Asn Ile Gly Tyr
275 280 285
Ser Glu Lys Asp Arg Phe Gln Gly Arg Phe Asp Val Lys Ile Glu
290 295 300
Val Lys Ser

- (2) INFORMATION FOR SEQ ID NO:9:
(i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 24 bases
 (B) TYPE: nucleic acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: linear
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:9:

ATGGGGAAGG GGGTTGGACG TGAT 24

- (2) INFORMATION FOR SEQ ID NO:10:
(i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 24 bases
 (B) TYPE: nucleic acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: linear
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:10:

ATAGTAGGTT TCCTTCTCCA CCA 24

- (2) INFORMATION FOR SEQ ID NO:11:
(i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 24 bases
 (B) TYPE: nucleic acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: linear
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:11:

ATGGCCCGCG GGAAAGCCAA GGAG 24

- (2) INFORMATION FOR SEQ ID NO:12:
(i) SEQUENCE CHARACTERISTICS:
 (A) LENGTH: 24 bases
 (B) TYPE: nucleic acid
 (C) STRANDEDNESS: single
 (D) TOPOLOGY: linear
(xi) SEQUENCE DESCRIPTION: SEQ ID NO:12:

GCTCTTAACT TCAATTTTTC CATC 24

- 59 -

WHAT IS CLAIMED IS:

1. A delivery system for delivering a therapeutically effective amount of a predetermined genetic material to myocardial cells of a chosen location of a patient's heart, said genetic material being selected for the function of increasing the amplitude of the patient's cardiac signal so that it can be better sensed by an electrode, comprising:
 - 10 a supply of said genetic material;
reservoir means for containing said genetic material; and
delivery means for delivering said genetic material from said reservoir to said myocardial cells,
 - 15 thereby increasing the amplitude of the cardiac signal and improving the signal to noise ratio that can be sensed by a pacemaker.
2. The delivery system of claim 1, wherein said supply of genetic material comprises a bolus of ion channel protein genetic material selected for the function of increasing the amplitude of the cardiac signal.
3. The delivery system of claim 1, wherein said delivery means comprises a catheter with a distal end portion, and said reservoir means is located in said distal end portion.
4. The delivery system of claim 3, wherein said distal end portion comprises a hollow helical element forming an interior, and said reservoir means comprises said interior with said supply therein.
5. The delivery system of claim 1, wherein said delivery means comprises a catheter with a lumen for delivering said genetic material therethrough, said catheter having a distal tip communicating with said lumen for

- 60 -

contacting said plurality of cells in the proximity of said electrode with said genetic material.

6. The delivery system of claim 5, wherein said distal tip is a hollow helical needle tip.

5 7. The delivery system of claim 5, wherein said catheter is a transvenous endocardial catheter.

8. The delivery system of claim 1, wherein said reservoir contains a supply of 0.1-10 ml of said genetic material.

10 9. The delivery system of claim 1, wherein said delivery means comprises a catheter with a distal portion and an end tip, and wherein said reservoir means is contained in said distal portion, and further comprising force means for forcing said genetic material from said
15 reservoir means and out of said end tip.

10. The delivery system of claim 9, wherein said force means comprises a stylet.

11. The delivery system of claim 1, wherein said delivery system comprises a hollow helical screw-in element
20 loaded with a bolus of said genetic material.

12. The delivery system of claim 11, wherein said element comprises ports for egress of said genetic material into said identified cardiac location when said element is screwed into said location, and further comprising soluble
25 plugs in said ports to maintain them normally closed but which dissolve when said element is positioned within said patient's heart.

13. The delivery system of claim 1, wherein said predetermined genetic material is DNA or RNA, and imparts

- 61 -

chronic change in ion channel expression in said cardiac cells.

14. The delivery system of claim 1, wherein said delivery means comprises a catheter with a distal end
5 portion, and said reservoir means is located in said distal end portion.

15. The delivery system of claim 13, wherein said DNA or RNA encodes an ion channel protein.

16. The delivery system of claim 15, wherein said
10 ion channel protein is a sodium channel protein.

17. The delivery system of claim 16, wherein said sodium channel protein is hH1.

18. The delivery system of claim 1, wherein said predetermined genetic material is protein, and imparts acute
15 change in sodium channel expression in said cardiac cells.

19. The delivery system of claim 18, wherein said protein is an ion channel protein.

20. The delivery system of claim 19, wherein said ion channel protein is a sodium channel protein.

21. The delivery system of claim 20, wherein said
20 sodium channel protein is hH1.

22. An implantable delivery system for delivering doses of a therapeutically effective amount of a predetermined genetic material to myocardial cells in a
25 chosen location of a patient's heart, comprising:

a supply of genetic material of the class having the property of increasing the expression of ion channels in the myocardial cells to which it is delivered;

- 62 -

a catheter, said catheter having a distal tip portion for engaging the cells of said chosen location and delivering thereto said genetic material;

reservoir means for holding said supply of genetic material and providing it to said distal tip portion of said catheter; and

delivery means for delivering a therapeutically effective amount of said genetic material from said reservoir means through said distal tip portion to said chosen location.

23. The system as described in claim 20, further comprising:

control means for controlling operation of said delivery means to deliver respective said doses.

24. The implantable delivery system of claim 23, wherein said control means comprises initiating means for initiating delivery of said genetic material, said initiating means comprising an external programmer.

25. The implantable delivery system of claim 23, wherein said control means comprises automatic means for automatically initiating delivery of said genetic material.

26. An implantable delivery system for delivering predetermined genetic material to cardiac cells adjacent to a pacing electrode positioned against the inner wall of a patient's heart, comprising:

a supply of genetic material of the class having the property of increasing the expression of ion channels in cardiac cells to which it is delivered;

a catheter, said catheter having a distal tip portion for engaging said cardiac cells and delivering thereto said genetic material;

- 63 -

reservoir means for holding said supply of genetic material and providing it to said distal tip portion of said catheter; and

delivery means for delivering a therapeutically effective amount of said genetic material from said reservoir means through said distal tip portion to said cardiac cells.

27. The implantable delivery system of claim 26, wherein the distal end of said distal tip portion further comprises a pacing electrode.

28. The system as described in claim 26, further comprising:

control means for controlling operation of said delivery means to deliver respective said doses.

29. The implantable delivery system of claim 26, wherein said control means comprises initiating means for initiating delivery of said genetic material, said initiating means comprising an external programmer.

30. The implantable delivery system of claim 26, wherein said control means comprises automatic means for automatically initiating delivery of said genetic material.

31. An implantable system for pacing a patient's heart and for delivering a predetermined genetic material to cardiac cells adjacent to a pacing electrode positioned in said patient's heart, comprising:

a supply of genetic material of the class having the property of increasing the expression of ion channels in cardiac cells to which it is delivered;

a catheter, said catheter having proximal and distal ends, a lumen through at least a part thereof and connecting to said distal end, a pacing electrode positioned at said distal end for engaging said patient's heart wall,

- 64 -

said electrode having a channel therethrough in communication with said lumen, and a conductor connecting said proximal end to said electrode,

5 a pulse generator connected electrically to said conductor at said catheter proximal end for delivering pace pulses to said electrode,

reservoir means for holding said supply of genetic material, and

10 delivery means for delivering said genetic material from said reservoir to said lumen, whereby said material passes through said lumen and said channel to said heart wall.

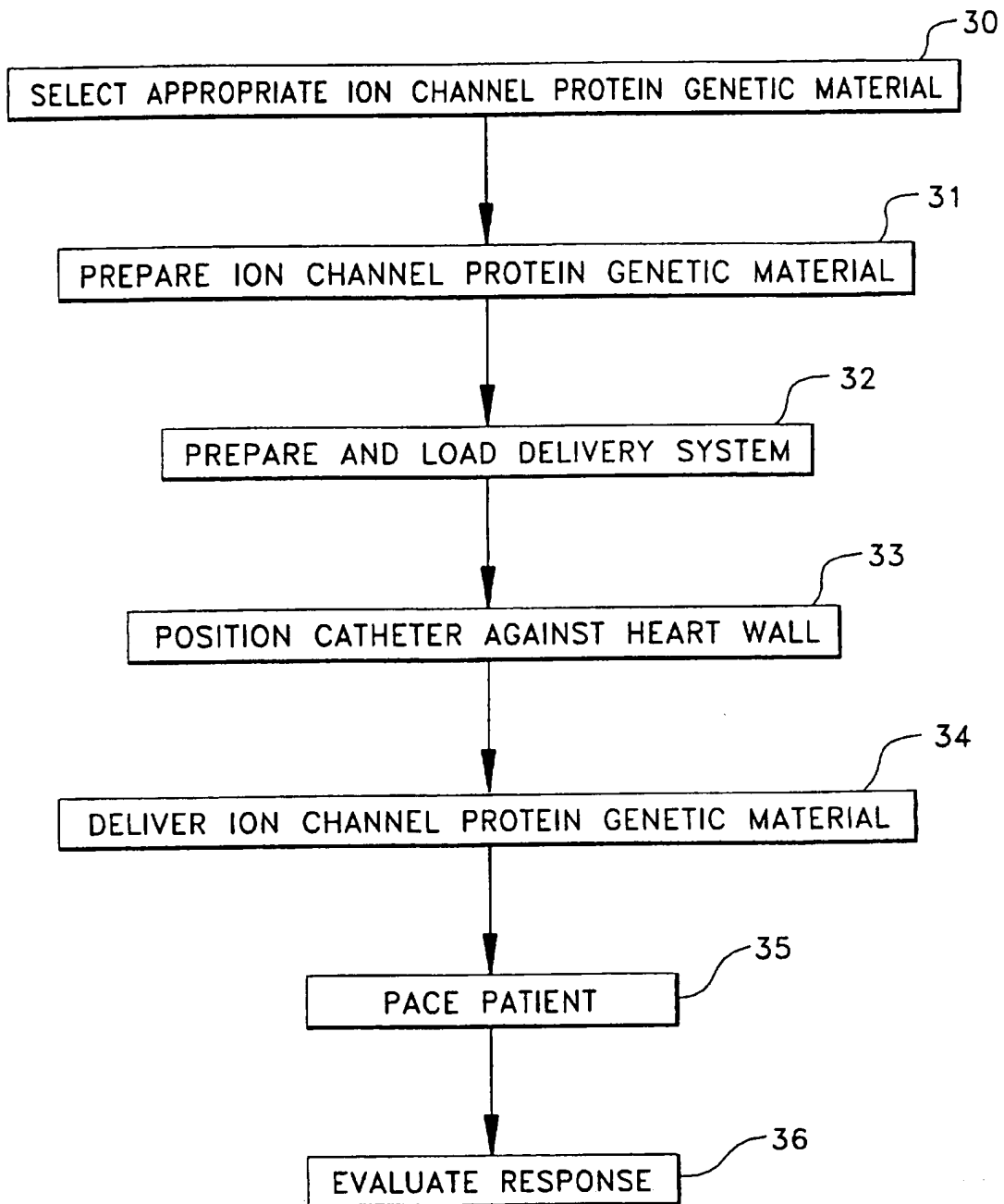
32. The implantable system of claim 31, wherein said reservoir is mounted in said pulse generator.

15 33. The implantable system of claim 31, wherein said delivery means is passive.

34. The implantable system of claim 31, wherein said delivery means comprises a pump.

20 35. The implantable system of claim 31, wherein said electrode is substantially concentric with respect to the catheter axis, and the channel passes through the center of said electrode.

1/5

*FIG. 1*

2/5

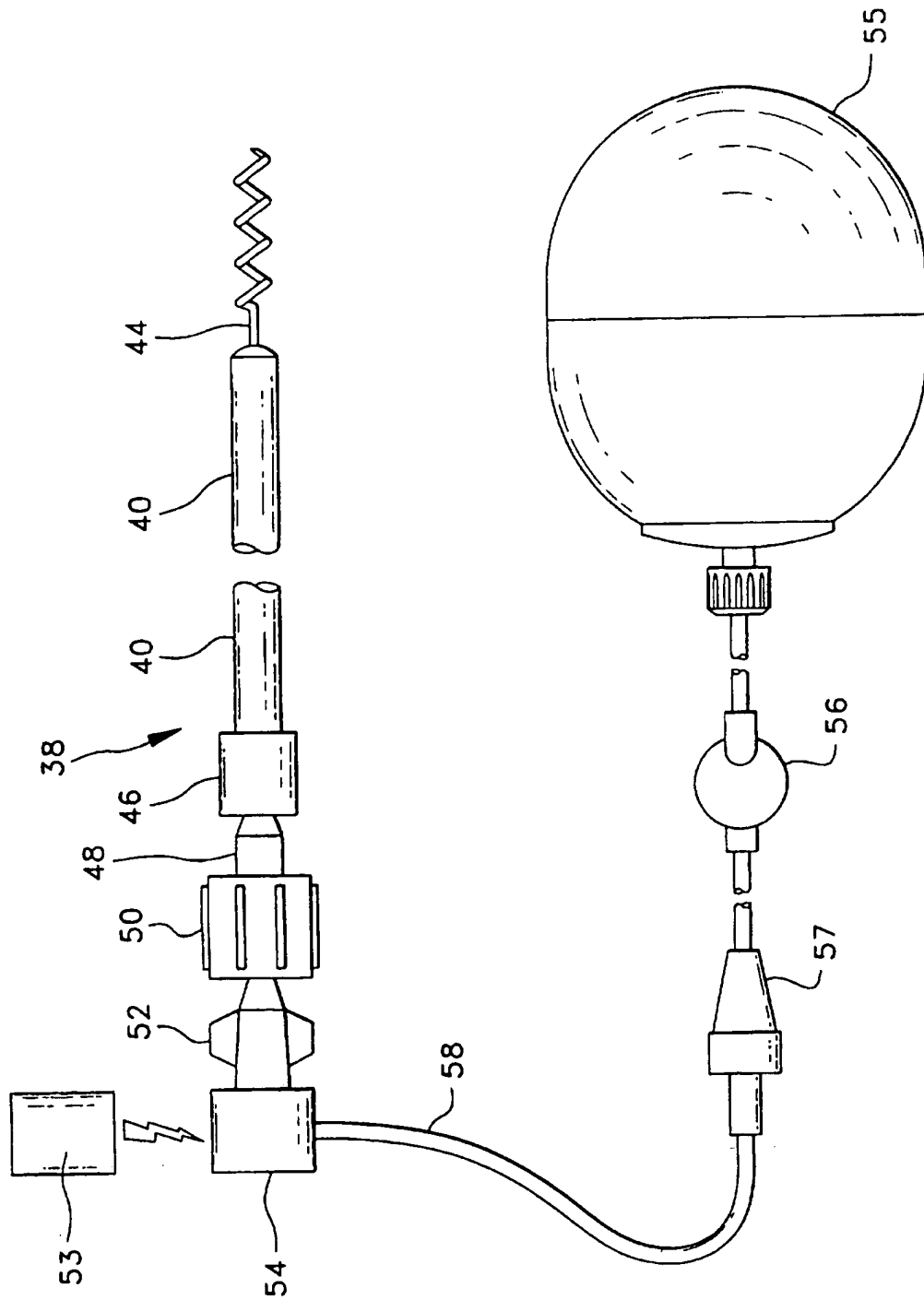


FIG. 2

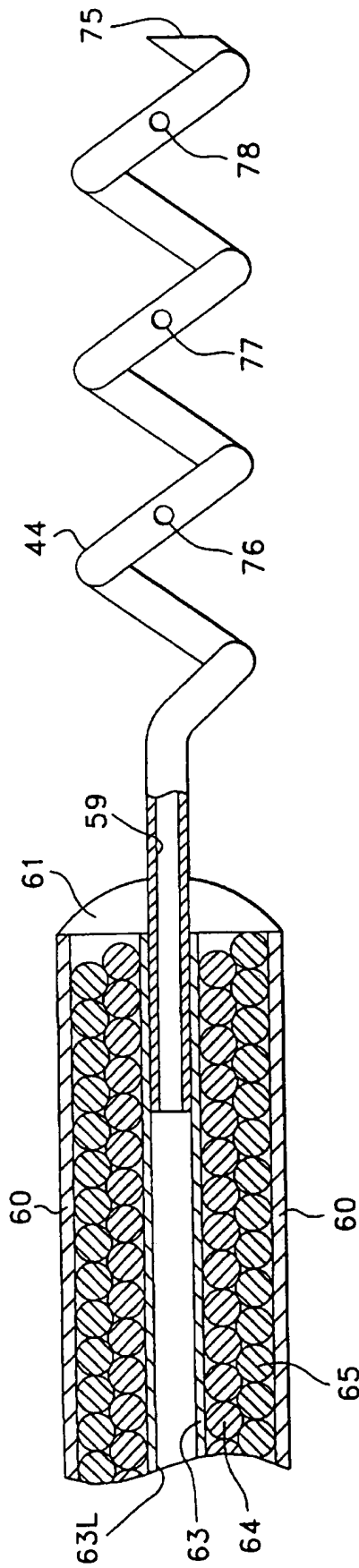


FIG. 3

3/5

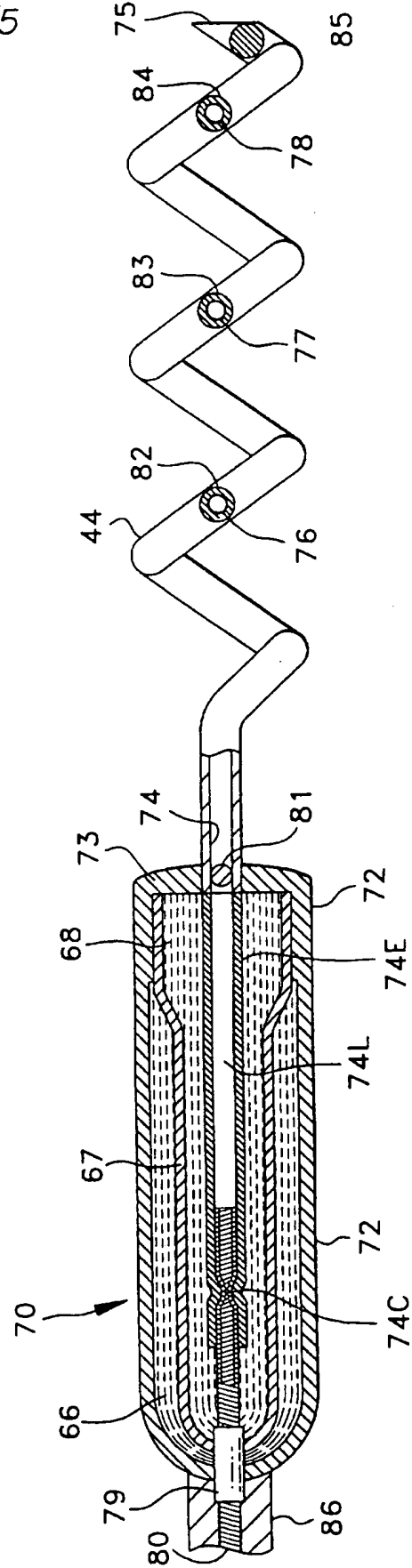


FIG. 4

4/5

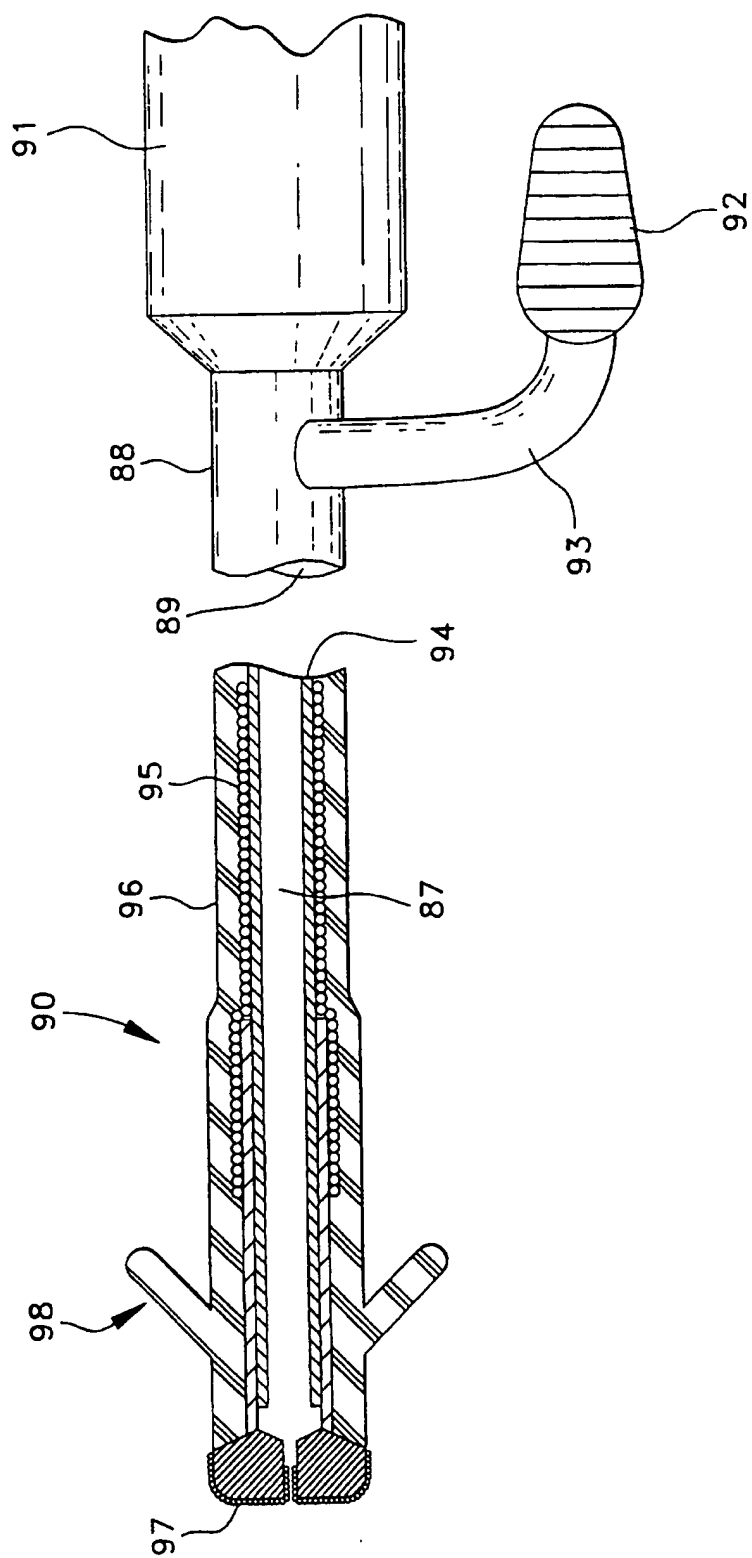


FIG. 5A

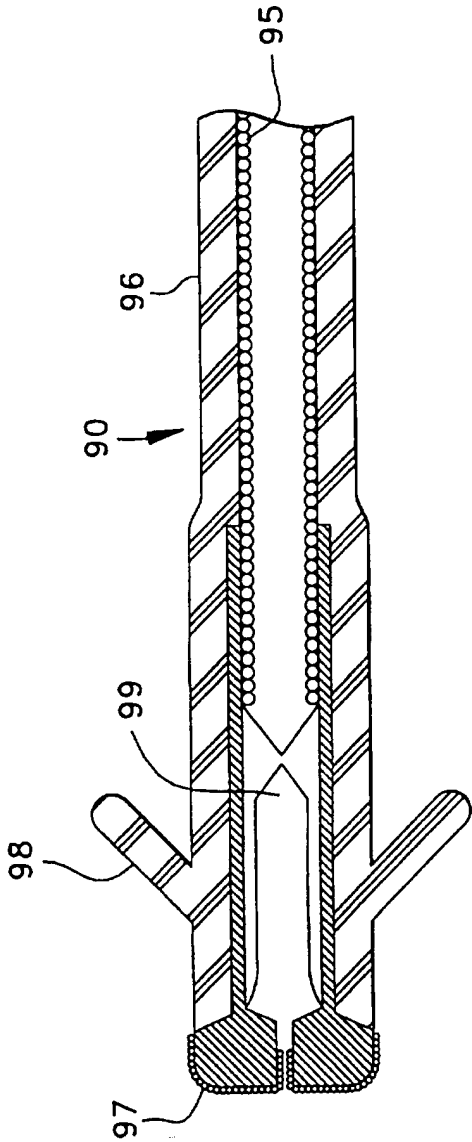


FIG. 5B

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US97/05556

A. CLASSIFICATION OF SUBJECT MATTER

IPC(6) : Please See Extra Sheet.

US CL : 514/44; 536/23.1; 435/320.1; 607/120

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 514/44; 536/23.1; 435/320.1; 607/120

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

DIALOG: MEDLINE, BIOSIS, EMBASE, DERWENT; APS

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5,496,360 A (D.A.HOFFMAN) 05 March 1996, see abstract	1-35
Y	US 4,711,251 (K.B. STOKES) 08 December 1987, see abstract	1-35
Y	NABEL et al. Recombinant Gene Expression in Vivo Within Endothelial Cells of the Arterial Wall. Science. Vol. 244, pages 1342-1344, see entire document.	1-35
Y	GELLENS et al. Primary structure and functional expression of the human cardiac tetrodotoxin-insensitive voltage-dependent sodium channel. Proc. Natl. Acad. Sci. USA. January 1992, Vol. 89, pages 554-558, see entire document.	1-35



Further documents are listed in the continuation of Box C.



See patent family annex.

* Special categories of cited documents:	*T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
A document defining the general state of the art which is not considered to be of particular relevance	*X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
E earlier document published on or after the international filing date	*Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
L document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	*G* document member of the same patent family
O document referring to an oral disclosure, use, exhibition or other means	
P document published prior to the international filing date but later than the priority date claimed	

Date of the actual completion of the international search

12 MAY 1997

Date of mailing of the international search report

12 JUN 1997

Name and mailing address of the ISA/US
Commissioner of Patents and Trademarks
Box PCT
Washington, D.C. 20231

Facsimile No. (703) 305-3230

Authorized officer

ABDUR HAZZAQUE
Telephone No. 703-308-0196

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US97/05556

A. CLASSIFICATION OF SUBJECT MATTER:

IPC (6):

A01N 43/04; A61K 31/70; C07H 21/02, 21/04; C12N 15/00, 15/09, 15/63, 15/70, 15/74; A61N 1/04